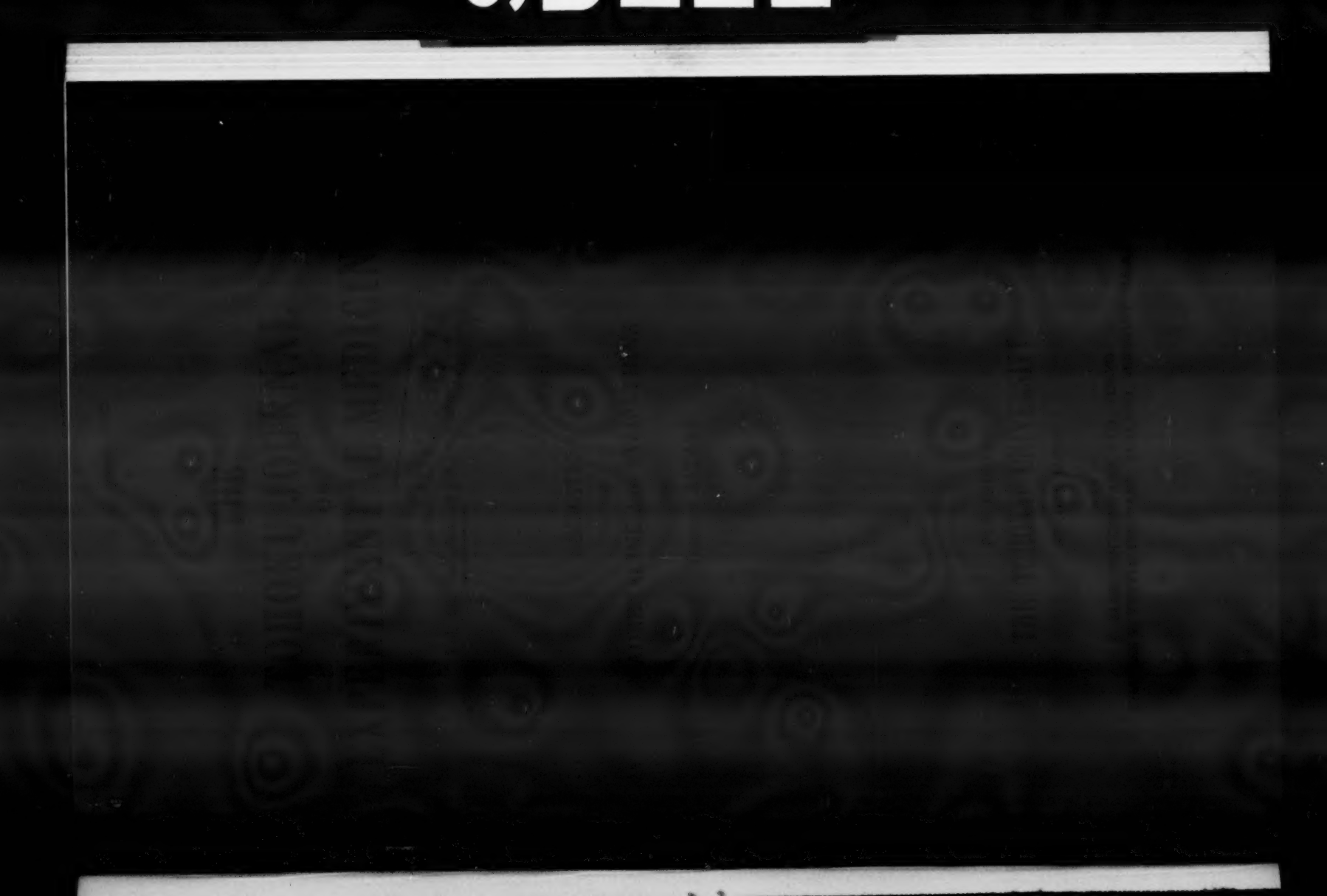

SECRETION OF ADRENALINE AND SYMPATHINS

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PREFACE

> "I often say that when you can measure what you are speaking about, and express it in numbers, you know something about it; but when you cannot measure it, when you cannot express it in numbers, your knowledge is of meagre and unsatisfactory kind: it may be the beginning of knowledge, but you have scarcely, in your thoughts, advanced to the stage of *science*, whatever the matter may be."
>
> William Thomson, 1883

ABOUT a decade after the isolation of adrenaline, the study of its secretion was begun, various methods being devised in that and the following decade. Those which were devised in the early period were of course carried out under artificial experimental conditions, so that a sound inference could be drawn only with insurmountable difficulties.

In the later half of the third decade the methods were elaborated to estimate the secretion of adrenaline alone or together with other sympathomimetic hormones in the animal without fastening, anesthetizing, laparotomizing or giving any pain. Of the adrenal blood collected under such conditions, adrenaline or sympathomimetic power is determinable in various ways, biologically and chemically as well. The outcome of auto-assay with a denervated organ is to be evaluated with certain qualifications, inherent to that organ.

Improved methods and technique provide the physiology of the secretion of adrenaline and the like some approaches in realizing its development towards a mathematical biology.

The quantitative data concerning the fluctuations in the adrenaline secretion on various stimulants in the almost normal dog, with simultaneous determination of some functions' variations associated with the former, and their correlation make up the chief content of this book (Chapters VII and VIII). This is supplemented by a study of the results witnessed in simultaneous use of the denervated organ with the lumbar route experiment. It sup-

plies a clue to estimate differentially sympathomimetic hormones besides the adrenaline.

As regards relevant literature, particularly for the last two chapters, only those having a precise and direct bearing upon the data there given are referred to. These writings and the others, quoted to show the general trend of matters, both mostly from this laboratory, involve the bibliography and discussion of them as thoroughly as possible up to the time of their publication. Since the War reference work is still difficult in this country.

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Further, I take this opportunity to acknowledge my indebtedness to Messrs. J. Madeley, P. Gerhard, G. Würfel, S. H. Griggs and other gentlemen who have kindly looked over our papers, written in English or German, through a long course of over thirty years, and on this occasion my thanks are gratefully given to Mr. S. H. Griggs for carefully looking over the present manuscript.

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CHAPTER I

INTRODUCTORY

SECRETION of adrenaline from the adrenal glands was first traced by a French physiologist as early as about a century ago.* But the matter was apparently most actively studied in the middle of the first half of this century when two competent physiologists, Cannon and Stewart, published their findings from a large number of experiments one after another, done with different methods, and in fact with quite opposite inferences. The one takes a denervated organ in the auto-assay as the indicator of the adrenaline secretion, and the other estimates the adrenaline in the blood taken from the adrenal gland under certain animal conditions. Concerning the former method, one should invariably keep in mind the logic that "the reverse is not always true", and further, that the latter method was carried out under conditions too artificial.

This period was then followed by that when an animal was experimented on without fastening, anesthetizing, laparotomizing, or evoking any pain, either on the dog, the adrenal blood of which was *seriatim* collected, or mainly on the cat with long-lasting preparation of a denervated organ or tissue. The latter indicates the combined secretion of the sympathomimetic hormones, and it is impossible with this method to determine directly the secretion of the adrenaline alone.

A long series of experiments was carried out by means of the former method on various kinds of subjects over a period of about a quarter of a century. All the data are here reviewed anew, discussed and systematically arranged in this book (Chapter VII and VIII).

However, a systematic study was done only with certain agents which were proven to accelerate copiously the adrenaline secretion: many other subjects do not act so strongly; intermediate and small doses were not tried with the former group of agents. The agencies tested were not always those which are ordinarily produced in normal life in the animal organism; many of them were pharmaceutic or the like. The results, therefore, fall far short of giving the whole story of the adrenaline secretion and its integral actions occurring in the animal individual, that is to say, its regulating actions on various organs and tissues sympathetically innervated.

Parallel with the determination of adrenaline secretion, the auto-assay by means of the denervated heart and pupil was carried out when cold was

* Vulpian, *C. r. Acad. Sci.*, 1856, **43**, 663.

applied to the animal body. This yielded a clue for acknowleding the course of the secretion of sympathins. A discussion of this will be found in Chapter VIII.

With elucidation from some hundreds of experiments, it is worth while mentioning here that the speed with which the blood flows out from the adrenal vein remains without much change. The variation is here only three times *ad maximum*, i.e. never in such large quantities as to vitiate very substantially the inferences made by some previous writers who estimated only adrenaline concentration in the adrenal blood but not the rate of flow, or that in the blood in the inferior cava near the entrance of the adrenal vein, although they were pointedly criticized by the Cleveland physiologist. And this finding opens the way for estimating the variation of the adrenaline secretion rate in the animal, or even in the human being, only by inserting a catheter into or near the adrenal vein in a suitable manner, without disturbing the body conditions, if any, other than those than the procedure causes.

In studies of secretion of adrenaline and other sympathomimetic hormones, nothing can be said with complete exactness until the time comes when the sympathomimetic hormones transported in the blood can be determined chemically or biologically. During the second World War and thereafter both methods have been enriched by new devices, but cannot yet pretend to completeness.

Advancement in the methods of determining adrenaline and the like in tissues, especially in the adrenal gland, precedes as a matter of course than in the blood, and in fact a great number of methods have been devised for the former purpose, while there are still only a few methods available, and these as yet not perfected, for estimating adrenaline in the blood.

Inasmuch as the determination of adrenaline in blood premises the adequacy of research on the adrenaline secretion, properties of the methods with which it was done will be discussed in detail.

The percentages of adrenaline or the sympathomimetic properties in the adrenal gland form a definite correlation in respect to various kinds of animals and various kinds of methods, both biological and chemical. The correlation is well established, there being a sufficient number of experiments to support it (Chapter II).

Because of existence of a close relationship with the main question at issue, the reports available in our time of the nor-adrenaline have been collected as thoroughly as possible and epitomized in the same manner as the cases with adrenaline (Chapter IV). The sympathin hypothesis cannot yet be wholly susbstituted for by the findings hitherto reported on the nor-adrenaline, and the designations such as sympathin, sympathin loc., sympathin remot., hepatic sympathin, etc. of Cannon, and adrenaline-intestine, adrenaline-pupil, etc. of ours can be used, even when the substances expressed

by the terms above named can be later completely chemically and particularly quantitatively analyzed. Only the hypothetical sympathins E and I can not be accepted in principle or in practice (Chapter III).

The configurations of the nervous system, central and peripheral, in regulating the secretion of the sympathomimetic hormones are to be formulated more subtlely than before (Chapter V).

An over-all view of the activities of the adrenaline and other sympathomimetic hormones in various conditions of the organism, indicates the important role they play in the adjustment of its behaviour, though inferior in some degree as to speed and nicety of time adjustment to that of the nervous system. On the contrary the action spreads widely and lasts long.

CHAPTER II

ADRENALINE CONTENT OF ADRENAL GLAND AND OF ITS MEDULLARY TISSUE*

CONTENT: On the evaluation of methods, chemical and biological, for measuring adrenaline in the adrenal gland—Adrenaline content of adrenal gland and its medullary tissue of various domestic animals—Is it reasonable to express the adrenaline content in terms of weight ratio whether of the gland or of animal body?—The cortex contains no adrenaline—From the radio of concentration of adrenaline reckoned on the gland weight and that on the medullary tissue, the ratio of weight of the gland and the medulla can be calculated, and *vice versa*—Relationship between adrenaline output and adrenaline content of the adrenal gland—Adrenaline diffuses from the medulla into the cortex after death of animal—No occurrence of compensatory increase in the adrenaline content of the one gland after removal of the other—"Virtual adrenaline"—Ascorbic acid in adrenal gland.

ADRENALINE content of the adrenal gland was measured most frequently for testing effect of a given agent thereupon; a reduction is commonly inferred as showing an augmented secretion of adrenaline, though not invariably correct.

A large number of methods, biological and chemical, were and are being devised for measuring adrenaline in the adrenaline gland; however the adrenal medulla, from a given species of animal yields varying content of adrenaline in general, depending upon the method applied, and *vice versa*. However, the numerals are in reality almost constant, and there are definite regularities and correlations among them.

While the adrenaline secretion has been much studied as applying to cats and dogs, the normal content of adrenaline in the adrenal gland has been estimated contrariwise only in a very small number of these animals.

Fresh, normal adrenals, which are clearly separable into the cortex and the medulla, and ample in quantity as to be determinable of adrenaline simultaneously by a large number of methods, chemical and biological, were taken from cattle, horses and pigs, killed in a slaughter-house. A certain number of normal dogs and rabbits were slaughtered in order to determine the normality. The extraction was made from " identically the same material " prepared by means of liquid air.

Inasmuch as uniform figures were obtainable from an adequate number of experiments, all the data are epitomized in Tab. I, and the materials thereof are given in detail in Tab. II; the data, not included in the tables, so far as considered sufficiently appreciable for comparison with these figures are

* All the data included in this chapter are those estimated before nor-adrenaline came to be investigated biologically. However, the data presented and discussed in this chapter will apply even after the time comes when all sympathomimetic principles in the adrenal glands can be quantitatively estimated.

referred to in the text, even though the experiments be small in number.

On the evaluation of methods, chemical and biological, for measuring adrenaline in the adrenal gland.

When a new chemical method of estimating adrenaline in the adrenal gland, or generally speaking, in animal tissue was devised, its evaluation was commonly tested by a parallel determination with biological methods, particularly so against the pressoric action method.

In the case of *Folin, Cannon and Denis' method*, Folin conducted chemical estimation and Cannon blood pressure experiment, independently of each other.[1] Both estimations gave wholly identical figures with sheep and cattle, they reported, and the figure from the cattle adrenal medulla well harmonizes with those presented in the accompanying Tbs. I & II. But the paper lacks biological estimates re the rabbit gland. A close agreement between both methods was once mentioned by Stewart and Rogoff in two experiments, presumably done on cats.[2] The Folin method cannot be used for the adrenal glands of all kinds of animals unconditionally; ascorbic acid is also determined by that method. That is the reason why the method yields an incomparatively large amount of adrenaline for rabbit adrenals compared with all the other methods (Cf. Tabs. I and II). This method was formerly much used in America and in our laboratory; the data reported therefrom, especially on rabbits, must be evaluated with due care. (Cf. Ascorbic acid in adrenal gland, on p. 14.)

The *iodine method* and the *ferric chloride method*, both rather recently elaborated by Euler yielded some applicable data.[3] His figures of the pressoric action of cattle adrenal medulla coincide with those in Tabs. 1 and 2, and also with those obtained by him with the ferric chloride method. While we have no experience of the latter, the figures of Euler with it, we note, are somewhat larger than those obtainable by other chemical methods. Adrenaline content of sheep adrenal, estimated by Abelous[4] with the iodine method wholly coincides with that of Battelli[5] with the ferric chloride method. Iodine method yielded not precisely identical figures with the pressoric method in the hands of Hale and Seidell,[6] but by modifying it Scoville came to find it of satisfactory compared with the biological test.[7]

In the hands of Barker, Eastland and Evers the *persulphate method* was proved as more satisfactory than the biological method, but the *molybdate method* gave incomparably high values.[8] The figures obtained by the former method from cattle, the medulla and the whole gland, are within the limits reported by us.[9] The findings of Devine[10] are entirely at variance with the views of Barker et al. about both methods.

The *Shaw's arsenomolybdic acid method* yields in the hands of the deviser

TABLE

Adrenaline Content Estimated

(in mg. per g.

	Blood pressure (dog)	Leg vessel (toad)	Intestine segment (rabbit)	Paradoxical pupil (cat)	Ear vessel (rabbit)
Cattle					
Whole gland	3.7 ±0.1	3.6 ±0.3*	4.5 ±0.3	3.0 ±0.1*	
Medulla	12.6 ±0.4	11.1 ±0.8	16.1 ±0.3	11.3 ±0.5	
Suto as 1	*1.35±0.05*	*1.22±0.06*	*1.68±0.05*	*1.18±0.05*	
Horse					
Whole gland	2.3 ±0.1	2.3 ±0.1	2.3 ±0.1	2.5 ±0.1	
Medulla	12.7 ±0.4	13.7 ±0.7	15.1 ±0.4	13.5 ±0.5	
Pig					
Whole gland	3.2 ±0.2	2.6 ±0.3	3.8 ±0.3	2.2 ±0.3	3.1 ±0.5
Medulla	13.1 ±1.0	10.9 ±1.1	18.5 ±1.4	10.4 ±0.9	13.1 ±2.0
Dog					
Medulla			16.4 ±0.5		
Rabbit					
Whole gland		0.71±0.02	0.48±0.02	0.59±0.02	0.73±0.03

* In all experiments given in the tables, adrenaline hydrochloride of Sankyo Co., The preparates used here were those from gland material; the synthetic preparation has

† Cf. ascorbic acid in the adrenal gland on p. 14.

himself figures that harmonize well with the pressoric action.[11] The values obtained from rabbits are small, but they are wholly within the limits reported by H. Sato.[12] The content noted for dogs is too small, about one-half of those given in Tabs. I and II. And the values of Shaw of dogs and rabbits are about 80% of those obtained by other methods is of highly noteworthy. The Shaw and the rabbit intestine gave a same value for rabbit adrenals. That of cattle adrenal medulla, estimated by Devine by means of the molybdate test is a little larger than those given in Tabs. I and II. The author noticed that ascorbic acid is also estimable by that method.[10]

Adrenaline content of adrenal gland and medullary tissue of domestic animals.

The data included in Tabs. I and II are those obtained from an adequate number of experiments which, in fact, were undertaken by two or three groups of investigators, separated by the elapse of some years. The figures in each set of experiment are almost the same among the groups, as given in Tab. II; this permits the epitomizing of them further in Tab. I, in which the numerals marked with an asterisk are those from the experiments carried out by a single investigator or group of investigators.

At the outset it may be remarked that the procedures of extracting in-

I

by Various Methods

of tissue)

Folin method	Suto mercury method	Stuber mercury method	Permanganate reaction (Zanfrognini)	Iodine method (Euler)	Persulphate reaction (Ewins)	Shaw arsenomolybdic acid method
3.4 ±0.2* 10.4 ±0.2 *1.03±0.01*	2.9 ±0.2* 10.0 ±0.2 *1*	10.8 ±0.2 *1.09±0.01*	9.6 ±0.3* *0.98±0.02*	10.1 ±0.3* *1.01±0.05*	9.4 ±0.3* *0.96±0.02*	10.2 ±0.2 *1.03±0.01*
2.8 ±0.2 11.8 ±0.2	2.2 ±0.3 10.5 ±0.2	11.5 ±0.2				10.3±0.2
2.5 ±0.1 7.9 ±0.3	1.8 ±0.2 8.4 ±0.5					
10.7 ±0.4	10.0 ±0.3	10.0 ±0.3				8.0 ±0.2
1.34±0.03†	0.65±0.02	0.65±0.02				0.52±0.03

assayed by means of Folin-Kodama (*J. Biochem.*, 1922, **1**, 281) was used as the standard. been produced there after the outbreak of the War.

cluded in the Folin, Suto and Shaw methods gave no differences at all to the results of the assays. The different figures obtained are due therefore only to different methods of determination, different materials, or both.

The numerals of Tab. I indicate the general trend at a glance. In detail: adrenal medulla of cattle contains adrenaline in 10–11%, generally speaking; the persulphate method gives a smaller value, and the rabbit intestine method a much greater one. More precisely said, the Suto method, the iodine method and the molybdate method yield quite the same value, but the manganate method and the Folin method give a little different value, while the Stuber method and all the biological methods other than the rabbit intestine segment method provide a higher value by ten percent.

Adrenal gland of the horse shows rather exceptionally, in contrast to other species of animals under question, a similar concentration of adrenaline regardless of different methods of estimation, chemical and biological, so far as the present methods are concerned, especially in the case of the whole gland. Even the rabbit intestine segment method yields also a value not very different.

Both methods, the Suto and the Stuber, rely upon the oxidative action of the mercury upon the adrenaline, and both methods give quite the same

TABLE

Adrenaline Content Estimated

(in mg. per g.

	Blood pressure (dog)	Leg vessel (toad)	Intestine segment (rabbit)	Paradoxical pupil (cat)	Ear vessel (rabbit)	Folin method	Suto mercury method
Cattle							
Whole gland	3.7 ±0.1 3.8 ±0.4	3.6 ±0.3	4.6 ±0.5 4.5 ±0.4	3.0 ±0.1		3.4 ±0.2	2.9 ±0.2
Medulla	12.4 ±0.8 11.1 ±0.5	11.1 ±0.8	15.5 ±0.7 15.1 ±0.6	10.7 ±0.7		9.6 ±0.4	9.0 ±0.5
Medulla	14.8 ±0.6		17.3 ±0.8	11.7 ±0.6		10.7 ±0.2	10.1 ±0.2
Medulla			15.8 ±1.1 16.9 ±0.9			10.2 ±0.5 10.5 ±0.9	9.9 ±0.4 10.0 ±0.3
Horse							
Whole gland	2.3 ±0.1 2.2 ±0.1	2.3 ±0.1	2.2 ±0.1 2.5 ±0.2	2.5 ±0.1		2.8 ±0.2	2.2 ±0.3
Medulla	13.8 ±0.9 12.5 ±0.4	13.7 ±0.7	15.1 ±1.4 16.0 ±1.1	13.5 ±0.5		12.8 ±0.6	11.7 ±0.6
Medulla			15.4 ±0.8 14.7 ±0.6			11.8 ±0.3 11.6 ±0.3	10.4 ±0.3 10.3 ±0.3
Pig							
Whole gland	3.2 ±0.2 3.1 ±0.3	2.6 ±0.3	3.8 ±0.3	2.2 ±0.3	3.1 ±0.5	2.5 ±0.1	1.8 ±0.2
Medulla	13.1 ±1.0	10.9 ±1.1	18.5 ±1.4	10.4 ±0.9	13.1 ±2.0	7.9 ±0.3	8.4 ±0.5
Dog							
Medulla			16.4 ±0.5			10.7 ±0.4	10.0 ±0.3
Rabbit							
Whole gland		0.71±0.02	0.46±0.02 0.50±0.03 0.54±0.04	0.59±0.02	0.73±0.03	1.32±0.03† 1.42±0.06 1.38±0.07	0.66±0.02* 0.64±0.03* 0.67±0.04

* Suto extract.

† Cf. Ascorbic acid in the adrenal gland on p. 13.

value of adrenaline concentration for the adrenal medulla of the dog and the whole gland of the rabbit. But the Stuber method supplies a higher value by just 8% than the Suto for the medullary tissue of cattle and the horse.

Shaw's molybdate method gives almost the same concentration of adrenaline as the Suto method for the medullary tissue of cattle and the horse on the one hand, and a smaller concentration of adrenaline by 20%, on the other hand, for the medullary tissue of the dog. The same is true for the whole gland of the rabbit. The Shaw method is noteworthy in these respects.

While the rabbit intestinal segment method gives a definitely greater value of adrenaline for the whole gland and the medulla of cattle and the horse in comparison with the other methods, chemical and biological, the adrenaline content, estimated by the intestinal assay for the whole gland of the horse,

II

by Various Methods

of tissue)

Stuber mercury method	Permanganate reaction (Zanfrongnini)	Iodine method (Euler)	Persulphate reaction (Ewins)	Shaw arsenomolybdic acid method	
					Folin extract / Suto extract } Kojima et al.[9]
					Folin extract / Suto extract } Kojima
	9.6 ±0.3	10.1 ±0.3	9.4±0.3		Folin extract / Suto extract } Hatano[13]
10.7 ±0.4 10.8 ±0.3				10.1 ±0.4 10.4 ±0.3	Folin extract / Shaw extract } Arai[14]
					Folin extract / Suto extract } Kojima
					Folin extract / Suto extract } Kojima
11.5 ±0.3 11.5 ±0.3				10.3 ±0.2 10.2 ±0.4	Folin extract / Shaw extract } Arai
					Folin extract / Suto extract } Kojima
					Folin extract / Suto extract } Kojima
10.0 ±0.03				8.0 ±0.2	Shaw extract-Arai
					Folin extract-Kojima
0.65±0.03 0.65±0.04				0.51±0.04 0.52±0.04	Folin extract / Shaw extract } Arai

harmonizes with that arrived at by other methods, a little smaller in some cases, the same or a little more in others; definitely smaller than the Folin method. Further, the intestinal method yields the smallest concentration of adrenaline for the whole gland of the rabbit, and almost the same as the molybdate test. An extraordinarily high value of the Folin in the case of the whole gland of the rabbit is due to the existence of ascorbic acid in the cortex.

Briefly summarized: provided about ten experiments, for example, be run, an almost constant figure is obtainable for each one of the determining methods of the adrenaline concentration in the whole gland and the medullary tissue of a species of animal. A glance at Tabs. I and II verifies that there are certain variations in the values obtainable by one and the same determining method according to the material from various kinds of animal and *vice versa*, and in certain cases there are remarkable regularities, as above

pointed out. These differences and these coincidences can never be too highly appreciated. The methods of extraction here applied have no significant influence upon the differences, here observed.

Various conditions which interfere with the determination of adrenaline in organ extract, etc. have been studied for long on the one hand;[15] on the other the existence of sympathomimetic hormones associated with the adrenaline has been discovered in the adrenal gland rather recently. Studies thereof proceed rapidly. Several methods of determination, chemical and biological, have been devised on these occassions. The data, the differences, and the coincidences here presented would be made clearly intelligible by further advancement along those lines of studies.

In such circumstances, we are still of opinion that it is practical and desirable to name the adrenaline in the adrenal gland, e.g., estimable by the use of the Suto method, the Shaw, the blood pressure, the rabbit intestine segment, etc., etc. as adrenaline-Suto, adrenaline-Shaw, adrenaline-blood pressure, adrenaline-intestine, etc., etc.

Is it reasonable to express the adrenaline content in terms of weight ratio whether of gland or of animal body?

Generalizing, the adrenaline content varies individually within about thirty per cent in all the animal species, and whether it be expressed in terms of per weight of the gland or of the body has no significant bearing upon the picture, as given in Tab. III.[16]

The rabbit makes an exceptional case;[12] the cortical mass is there abundant, twenty times that of the medullary, and varies from case to case. Accessory cortical tissue exists invariably in this species of animal. Ascorbic acid contained in the cortical tissue is also Folin-positive.

The above is said of normal animals. Under certain circumstances the three factors determining these ratios, i.e. the total amount of adrenaline, the gland weight and the body weight may alter. Experimental data in rabbits with diphtheria toxin,[17] physostigmine[18] and inanition[19] affords illustration thereof. Further, in comparing the adrenaline content in the fellow glands, one of which is extirpated before a certain length of time, comparison should be made with the amount of adrenaline per gland and per unit of body weight.[20]

The adrenal cortex contains no adrenaline.

It is possible to isolate the cortical tissue which does not contain any trace of the adrenaline from the medulla. An absolute clean separation of each type of tissue by dissection was accomplished in small animals as rabbits[21] and cats.[22] Similar data were also observed in other species of domestic

TABLE III

Showing the Adrenaline Content in Terms of Unit of the Gland or that of Body

Adrenaline content is indicated in mg., the gland weight in g. and the body weight in kg. The width of variation is expressed in percentage. M: Mathematical mean. α: Standard deviation. V: Variation coefficient.

		By the Suto method						By the Folin method					
		Adr./Gl.			Adr./B.W.			Adr./Gl.			Adr./B.W.		
		M	α	V	M	α	V	M	α	V	M	α	V
Kojima et al.	Cattle (10)	2.9	0.73	25%	0.086	0.025	30%	3.4	0.73	22%	0.101	0.032	31%
	Horse (6)	2.2	0.34	16%	0.106	0.022	21%	2.8	0.56	20%	0.136	0.030	22%
	Pig (10)	1.8	0.63	35%	0.050	0.016	32%	2.5	0.41	16%	0.15	0.06	42%
Sato-Rabbit (617)		0.56	0.22	40%	0.088	0.021	24%						
Fujii-Rabbit (148 male)								1.13	0.33	28%	0.237	0.087	31%
Fujii-Dog (6)								1.8	0.44	25%	0.22	0.051	24%
Kuriyama-Rat (20)								2.2	0.58	27%	0.043	0.014	32%

By the iron chloride method

		Adr./Gl.			Adr./B.W.		
		M	α	V	M	α	V
Battelli	Dog (5)	1.03	0.34	33%	0.082	0.020	25%
	Sheep (2)	1.45	0.07	4.8%	0.118	0.004	3.6%
	Pig (2)	1.80	0.07	3.8%	0.081	0.004	5.4%
	Cattle (2)	1.7	0.0	0	0.076	0.003	3.5%
	Horse (4)	1.10	0.24	22%	0.97	0.16	1.7%

animals, such as cattle, horse, sheep, etc.[23]

A histological study of Szent-Györgyi, in which the gland is immersed in 5% potassium iodate solution containing 30% acetic acid, shows that within 30 minutes the medulla turns dark violet, while the cortex remains unaltered in colour, proving also non-existence of adrenaline in the cortex.[24]

The findings[25] that each of both tissues, cortical and medullary, exists also in another layer, though very little, must also be kept in mind in such kinds of experimentation as the above.

From the ratio of concentration of adrenaline reckoned on the gland weight and that on the medullary tissue, the ratio of weight of the gland and of the medulla can be calculated, and vice versa.

The above statement, the subheading shows, is based on the fact that the cortex contains no adrenaline. This reckoning sometimes affords useful figures.

In the dog, for example, adrenaline-Suto exists in 1% in the whole gland, and in 10% in the medullary tissue; therefrom the ratio of the weight of the gland and of the medulla is to be assumed as being 10 : 1. If adrenaline-Folin be taken into account, it is 7 : 1 from the data as 1.7% against 11%.

The figures nearly harmonize with the finding of Elliott and Tuckett[26] who fixed adrenals with the mixture of $K_2Cr_2O_7$ with formalin, frozen and sectioned flatly; a ratio of the weight of the whole gland of the adult dog to the medulla is given then as about 7 : 1. Here such chemicals, as $AgNO_3$, KJ, can be also applied.

The data of Flint,[25] who hardened the dog gland with formalin, are decidedly different therefrom: 3.5 : 1 by weight and 3 : 1 by volume.

As a further step we may here try to find by reckoning in this manner the adrenaline content in the rabbit's adrenal medulla, since there is still no direct estimation of this. The whole gland contains adrenaline-Suto in 0.65% (Tabs. I & II). Elliott et al.[26] estimated the ratio of cortex : medulla as 40 : 1 for a rabbit weighing 3 kg. and as 20 : 1 for that of a 2 kg. rabbit (p. 339). This means that the medulla of the rabbit of 3 kg. (p. 341) contains adrenaline in 27% and that of a 2 kg. rabbit 14%. The body weight of the rabbits in our laboratory is 1.5–2 kg.

Accordingly the medulla of rabbits contains a somewhat larger amount of adrenaline, but comparatively not very much different from other kinds of animals. If the medulla of rabbits be assumed as to contain adrenaline in quite the same concentration as in other species of animals, the ratio of cortex : medulla by weight would be 15 : 1. The shadow of chromaffine substance in the rabbit adrenal, which was stained in the Kohn's solution and then made transparent according to the method of Spalteholz, does not indicate a large disproportion in weight between both layers.[27] The rabbits used in Fujii's work were 1.5 kg. or thereabouts in the majority of cases.

Relationship between adrenaline output and adrenaline content of the adrenal gland.

When the adrenaline rapidly and abundantly discharges from the adrenals, the content in the latter also diminshes. When the content be found to be smaller than the standard, it is most possibly due to an excessive discharge from the gland. But the reverse does not hold true invariably: it depends on the other hand upon a new production of adrenaline. Although it is true that adrenaline output takes place even after cutting out the splanchnic nerves, as in the nicotine poisoning, it is also true even in taking out the nerves completely, when the output be not markedly rapid, but the animal becomes only moribund, as in diphtheria toxin poisoning.

TABLE IV

Relation between the adrenaline output and the adrenaline content of the adrenal gland, left behind

	Output (per hr.)		Adrenaline in gland		
			total	per gland	per body weight
Normal 6 animals (Fujii[25])	0.1 mg.		1.7 mg. (0.9–3.7 mg.)	1.8‰ (1.3–2.3‰)	0.21‰ (0.15–0.27‰)
No narcosis, cava pocket, sensory stim. 11 examples (Kodama[29])	0.45 mg. (0.04–1.1 mg.)	90 min (70–135 min.)	1.3 mg. (0.8–1.7 mg.)	1.4‰ (1–1.8‰)	0.17‰ (0.1 –0.22‰)
Narcosis, cava pocket, sensory stim. 18 examples (Kodama[30])	1 mg. (0.1–2.3 mg.)	120 min. (75–160 min.)	1.0 mg. (0.5–2.9 mg.)	1.0‰ (0.6–2.4‰)	0.13‰ (0.06–0.4‰)

Some agencies were proved to act directly upon the adrenals, so as to cause a hyperglycemia there, as diphtheria toxin,[31] coldness,[32,33] physostigmine,[34] acetylcholine,[35] etc.

The recovery of adrenaline content there in the adrenal capsule is independent of whether the gland has its innervation or not.[36]

Adrenaline diffuses from the medulla in the cortex after death of animal.

After the killing of the animal, adrenaline diffuses from the medulla towards the cortex, when the separation is conducted between both layers later, the concentration of adrenaline, e.g. determinable by the rabbit intestine segment method, is found to be greater. Simultaneously the Folin value increases too, but this is in reality due solely to the diffusion of adrenaline. Kneading accelerates the diffusion.[37]

In their time Oliver and Schäfer sometimes obtained a very small pressoric effect from extract of cortex, which they properly explained by the post-mortem diffusion of medullary juice or other accidental contamination.[37]

No occurrence of compensatory increase in the adrenaline content of one adrenal gland after removal of the other.

No appreciable increase occurs, generally speaking, in the adrenaline content in the gland left behind for a long time after removal of the fellow gland, at least in rabbits.[22] By way of precaution it should be noticed here that all the procedures as regards technique in the extirpation of the gland must be quite the same both times, viz., at the first and the second extirpa-

tion.[26] The Folin method can give another answer to this kind of question.

" *Virtual adrenaline* ".

Half a century ago some French investigators saw an increase of adrenaline in the adrenal gland that was reserved for some length of time after removal of the gland. In explaining this fact they assumed there to be the existence of " virtual adrenaline ", a parent substance or adrenaline in combination.[39]

Their data, however, were discordant: variation existed among the investigators findings, and some readers declined to accept their view. Cf. (39)

In the hands of Ooisi, the adrenaline (Suto) in pig adrenal increases by about ten per cent in 24 hours in a vacuum excitator arranged upon sulphur. In twelve experiments the outcome was fairly uniform.[40]

Similar figures are given in Ishibashi[41] and in Schild.[42] The procedures of the latter are not quite the same as those of Ooisi, and no word is said of the virtual adrenaline; but it may not be out of place to quote it here. Euler and Hamburg take the results as indicating the existence of sympathomimetic compound other than the adrenalin in adrenal extract.[43]

Ascorbic acid in the adrenal gland.

The phosphor tungstic method of Folin, Cannon and Denis gives a positive reaction against the ascorbic acid contained in the cortex. The Folin reaction in the cortex is due solely, highly probably, to that substance.

The content of ascorbic acid in the cortex tissue of various animals might be taken generally as 0.5 mg. per g. tissue. The mean for each species of animals is 0.3–1 mg. per g. tissue. The yields, obtained by means of the Folin methods are:

Rabbits (7 animals) 1 mg. per g. (0.7—1.5 mg.),[20] cattle (10) 0.3 mg. (0.1—0.7 mg.)[9] horses (6) 0.5 mg. (0.2—0.8 mg.),[9] cats (11) 0.5 mg. (0.3—0.7 mg.).[21]

The above figures can be used practically to express them in the term of ascorbic acid. Its molecular weight is almost the same as that of adrenaline.

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CHAPTER III

ON SYMPATHINS

THE work of Cannon on one side and that of Stewart on the other made clear about three decades ago that there are some agencies that act to accelerate or modify the function of the denervated organ, besides the acceleration of adrenaline output from the adrenals at the same time, roughly speaking. Cannon named the active principle first the "sympathin", the mediator being actually the adrenaline locally set free, i.e. having a different source from that of the adrenaline.[1]

In advancing experimental researches, discovery was made by Cannon and others of sympathin having features other than those in adrenaline. Of the development of the "sympathin theory" there are many matters that could be taken up and discussed, but to do so would be irrelevant to our purpose here. Here this finding alone may be noted, that in his later years of investigation Cannon, with Lissák, expressed his view as follows: adrenaline is liberated at the endings of adrenergic fibres, and sympathin, which escapes from the stimulated region into the blood stream, is this adrenaline modified in the affected cells.[2] (Cannon and Lissák, p. 777.)

Of the sympathin in general, we shall put forward our opinions in more detail later (Chapter VIII), but in passing we may mention that we are now-a-days of the opinion that several figures of sympathins must be collected on one side, and various experiences about nor-adrenaline or the like must be obtained. Of the latter we are getting various new reports one after another.

Explaining how adrenaline acts to inhibit, for example, the intestinal peristalsis, Cannon postulated two kinds of sympathin, excitatory and inhibitory, i.e. E. & I. Specifically stated, there are inhibitory hormones for the intestinal muscles and excitatory hormones for the intestinal blood vessels, the former more powerful than the latter. This further development in the "Cannon theory" was not supported in general by subsequent writers.[3]

On this point we are still of the opinion that the intestinal relaxation brought about by stimulating the splanchnic nerve or by applying the adrenaline is explainable only by assuming the mechanism which exists in the intestine itself, which is inhibited by the agent now applied there. One and the same agent acts to contract, for example, the heart muscle by stimulating it, and to inhibit the intestinal muscle by inhibiting it. No further advancement after Langley's period of the receptive substance.[4]

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CHAPTER IV

ON NORADRENALINE

In following the synthetization of adrenaline, noradrenaline or arterenol was synthetized about half a century ago, but it was about a decade ago that it came to be studied as a hormone widely and actively.

Noradrenaline is commonly contained in the adrenals and the tissues innervated adrenergically, and was isolated in the adrenals and its preparation.[1] It has a very intimate concern with matters dealt with in this book. Study on this matter is now developing rapidly and will be found to have ample consideration with the question at issue. All our studies presented in other chapters, were carried out without having an exact and full knowledge of noradrenaline and the like, and are given now without special attention thereupon. It is known that in delving into the secretion of adrenaline and the like, the matter of noradrenaline must be taken as being present at the same time. In this book all the sympathomimetic power or the substance which is designated as adrenaline is taken simply as adrenaline, which, in fact may involves noradrenaline and other sympathomimetic substances besides the adrenaline itself.

Although this might not be the proper place to make precise pronouncements about noradrenaline, it is nevertheless, very significantly related in the long run to the query dealt with in this book. It is nowadays very eagerly studied, and the time when adrenaline, noradrenaline and the like are studied simultaneously will soon be here. We see already such studies, but they are not yet amply evaluated. In order to show what is involved in the matters presented and discussed in the following chapters, recent studies of noradrenaline will be briefly given here. The materials given are those gathered before we had become acquainted with recent studies of noradrenaline.

Several degrees of actions of noradrenaline upon various organs and tissues of various kinds of animals have been studied in many ways. For example, a demonstration revealed that the activity ratio of L.-noradrenaline to L.-adrenaline varies from 0.73 to 1, as the mean, when tested on the cat paradoxical pupil reaction, 2.1 : 1 on the dog blood pressure, and 2.8 : 1 on the rabbit intestine segment.[2] Further, the minimum effective dose for the rat uterus was determined as 0.5×10^{-6} g. for adrenaline, and as $75\text{–}300 \times 10^{-6}$ g. for L.-noradrenaline, and that for the cat nictitating membrane as $10\text{–}50 \times 10^{-6}$ g. and 1×10^{-6} g. respectively.[3]

However, much endeavour is still needed to extend researches along these

lines.*

Here it may be noted that noradrenaline has a much smaller action on the carbohydrate metabolism, being only one-tenth (1 : 8–20) that of adrenaline.[4] There are numerous studies related to the interrelationship between adrenaline oversecretion and the blood sugar increase, needless to say.

The content of noradrenaline in the adrenals in various kinds of animals may be arranged as in the accompanying table to illustrate this.

Of each experiment a certain number of them are needed in order to show the general trend.[5] In these examples the noradrenaline was determined at the same time with the adrenaline by means of the permanganate reagents.[6]

TABLE V

Noradrenaline Content of the Adrenal Gland of Some Animals, Reckoned on the Whole Sympathomimetic Power

(%)

Cattle (32 medullae)	Horse (4 medullae)	Pig (11 pigs)	Goat (15 goats)	Dog (15 dogs)	Cat (7 cats)	Rabbit (6 rabbits)
29	18.5	42	36.5	18.5	45	1.8
(14–39)	(12–26)	(30–52)	(21–47)	(7–29)	(38–53)	(0–11)

The absolute amount of adrenaline plus noradrenaline will coincide with those given in Tab. I & II, on p. *6–9* of this book ; for example, the amount of adrenaline in cattle medulla by the permanganate method and the Suto method are 9.6 mg. per g. gland, and 10 mg. per g. gland there respectively, and that for this determination is 9.92 mg. per g. gland ;[7] that for the horse adrenal medulla the Suto was 10.5 mg. per g. medulla, and that by the permanganate this time 10.65 mg. per g. medulla ;[7] for the pig adrenal by means

* Physiological or pharmacological actions of noradrenaline are mostly weaker than those of adrenaline (Toxicity,[a] intestine,[b] hen's rectal caecum,[c] uterus,[d] pupil,[e] nictitating membrane,[f] spleen,[g] etc.), generally speaking. On the contrary the pressoric action is greater than that of noradrenaline.[h] The reversal of the adrenaline reaction by ergotoxine and ergotamine does not occur by noradrenaline, as all the writers agree.[i] The studies of yohimbin,[j] cocaine[k] against the pressoric action do not agree with each other. a Biberfeld (1906, 1/2–1/3), Schultz (1909) ; b Greer et al. (1938), Tainter et al. (1948, L-Adr.=L-Noradr.), Holtz and Schümann (1949, Noradrenaline=1/5 Adrenaline), Holton (1949, equal), Bülbring and Burn (1949, intestine, stimulated by acetylcholine, equally sensitive) ; c Euler (1948, 10–25–50 : 1) ; d Barger and Dale (1910) ; Greer et al. (1938), Tainter et al. (1948), Holton (1949), Bülbring & Burn (1949) ; e Bacq and Frederique, (1935), Greer et al. (1938) ; f Greer et al. (1938), Bülbring & Burn (1949) ; g Graham (1949) ; h= : Biberfeld (1906), Schultz (1909), Holtz & Schümann (1949), Holton (1949), Noradrenaline>Adrenaline : Barger & Dale (1910), Green et al. (1938), Tainter & Tuller (1948), Euler (1948), Barcroft & Konzett (1949), Noradrenaline<Adrenaline : Holton (1949) ; i Barger & Dale (1910), Melvills (1937), Stehle & Ellsworth (1937), Greer et al. (1938), Tainter (1948), Euler (1948) ; j Raymont & Hamet (1930), Greer et al. (1938) ; k Raymont & Hamet (1930), Melvills (1937), Tainter (1948).

of the Suto method it was 1.8 mg. per g. gland there, and for this assay 1.75 mg. per g. gland.[7] For the dog adrenal the content of the Folin was 1.2–2.3 mg. per g. gland[8] and that in this determination 1.07. But there is a big difference in the rabbit adrenal. For the Suto method the result showed 0.65 mg. per g. gland and 0.338 mg. per g.[7]

Noradrenaline is found there in about 10–50 per cent, mostly in 20–40% of the total quantity of the whole sympathomimetic hormones in the adrenal gland.[9] Such somewhat great variations, as from 0 to 36%, was noted in preparations from glands too.[10] Rabbits have none or only a small amount of noradrenaline in the adrenal gland.[11] (In the experiments of rabbits in the above table, only one rabbit had noradrenaline in 11%).

Some adrenals were found to have no noradrenaline on the one hand, as in dogs (Bülbring and Burn: 0 in two glands and 14, 20, & 50% in three glands[12]), cat,[13] guinea-pig[14] and rat.[15] On the other hand 80% was found in a cat.[16]

Whales have a large quantity of noradrenaline, such as five times as much as adrenaline.[16] This is taken as being due to their abnorm conditions; for example, the gland might be exhausted of adrenaline stock by the central stimulation. Human pheochromocytoma has as much noradrenaline as 50 to 90 per cent.[17]

There are only few reports on the noradrenaline content of adrenals in human beings: data we have seen give it as about ten or more per cent of the total sympathomimetic substances.[18] Noradrenaline in Ozaki was 0.12 mg. per g. of gland, about fourhold of another paper (S. & W.).

While it is thought noradrenaline is merely a precursor of adrenaline and serves no independent function, there are some opinions that the normal reflex vascular effects are affected chiefly by noradrenaline.[19] Hydroxytyramine can be seen also in the adrenals.[20]

Influence of insulin, morphine, β-tetra or of splanchnic stimulation, etc. upon the discharge of noradrenaline from the adrenal, etc. was not extensively discussed.[21]

Lastly it may be said that L.-noradrenaline is suggested as the sympathin E, for example, detected on stimulation of the hepatic nerves.[22]

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CHAPTER V

METHODS BY WHICH SECRETION OF ADRENALINE AND THE LIKE CAN BE ESTIMATED QUANTITATIVELY AND OPPORTUNELY IN THE PHYSIOLOGICALLY NORMAL DOG

In order to estimate the secretion of adrenaline from the adrenal, quantitatively and opportunely, the quantity of blood flowing from the adrenal and the adrenaline content therein must be estimated from time to time. The general blood contains sympathomimetic hormon properties other than those in the adrenals too; for example, the lasting preparation of the totally denervated heart determines jointly the amount of adrenaline and sympathin-liver, etc. Moreover, it is now established that noradrenaline is present also in the blood from the adrenal gland. The subject matter and data presented in this book have an intimate concern with such facts, but our experiments and investigations were carried out before those facts had been established. The adrenaline dealt with in this book, is here simply presented as adrenaline itself, but in fact it involves the noradrenaline too.

In studying the significance of the secretion of adrenaline from the adrenals, one must acknowledge how it is secreted when the animal is quite calm, then next how it alters when the animal be experimented on in differing ways. To do this, an adrenal vein is reached while the dog behaves quite normal physiologically, and the blood therefrom is obtained from time to time; then, after having given the animal a certain agent the collection of blood is repeated.

In order to carry out an experiment on a dog while it behaves itself quite normally, one must previously de-afferented its operation field. The procedure is as follows: the posterior roots from the thoracic 10. to the lumbar 3. are cut off, and after a recovery from the operation, usually a matter of one month, an actual experiment may then be carried out. The dog is not fastened on the table, nor narcotized at all. It stands, lays down or sleeps at will on the table. On making an operation aseptically, the first lumbar vein is reached, the branches between it and the adrenal gland are properly preparated, ligated, and cut off between the two ligatures.

A cannula is put into the lumboadrenal vein, then a small bulldog clamp is laid on, and a somewhat strong silk thread is passed round. When wanted, the thread loop is drawn out and the bulldog clamp removed; then adrenal blood runs out from the free end of the cannula and is collected by means

of a cylinder. During experimentation the dog behaves quite normal; it takes biscuit, milk, etc. or sleeps.[1]

Although this method allows one to see the secretion of adrenaline as natural as possible, there still exist some limitations; for example, it is impossible to start the dog, thus operated on and adjusted on the table or in the room, to quarrel with another dog or a cat. However, that non-fastening of the animal, deprivation of the use of anesthesia (by applying the denervation of the operation field), and operations such as laparotomy, etc. considerably improve experiments on the adrenaline secretion has been definitely proved. For example, that the adrenaline secretion is almost the same whether it is estimated on adrenal vein blood collected without opening the abdomen or after opening it, is true only when sufficiently deep anesthesia is relied upon.[2] Preparing the cava pocket in the non-anesthetized, de-afferented, but fastened dog brings about a remarkable exaggeration of the adrenaline secretion in contrast to the rate yielded by means of the lumbar route preparation.[3]

What influence the de-afferentation of the adrenal region has upon the blood flow through the gland, has not yet been worked out with definiteness. The experiments with morphine, and insulin under avertin narcosis are not taken to be such instances.[4] The contention that the secretory nerves of the adrenals are involved in the posterior roots is erroneous, as is verified in the next chapter.

The lumbar route method on the de-afferented dog is shown partly by the accompanying Fig. 1. The blood from an adrenal gland is collected while the dog stands, sits or sleeps on the table. The animal, a quite healthy one, is not fastened, narcotized, or laparotomized and no pain is evoked. The blood samples taken are to be estimated of adrenaline by certain methods, here, for example, by means of the rabbit intestine segment method, and sometimes simultaneously with the cat paradoxical pupil method.

(A) Normal Rate of Venous Blood-Flow through the Adrenal Gland of the Dog

In the experiments (268 in toto) reported between 1927–1940, the rate enumerated in one gland was 0.1–0.7 cc. per kg.-min., and in both gland combined 0.6 cc. per kg.-min. In detail: total (268 cases) 0.31 cc.$\pm$0.007; L. adrenal gland (201 cases) p.30 cc.$\pm$0.007, R. (67 cases) 0.33 cc.$\pm$0.017; males (180 cases) 0.29 cc.$\pm$0.07, females (88 cases) 0.35 cc.$\pm$0.014. That is to say, the blood-flow through the adrenal gland is more rapid in female dogs than in males—say, by 10 to 20%. The right gland has a little greater blood-flow than the left, though insignificant.

In about thirty dogs, the gland was denervated 2–4.5 hours before starting collection of adrenal blood, i.e. the splanchnic nerves were cut through. The blood-flow was reduced by one-third. (In 29 dogs, the rate was 0.21 cc.$\pm$0.017 per kg.-min.)

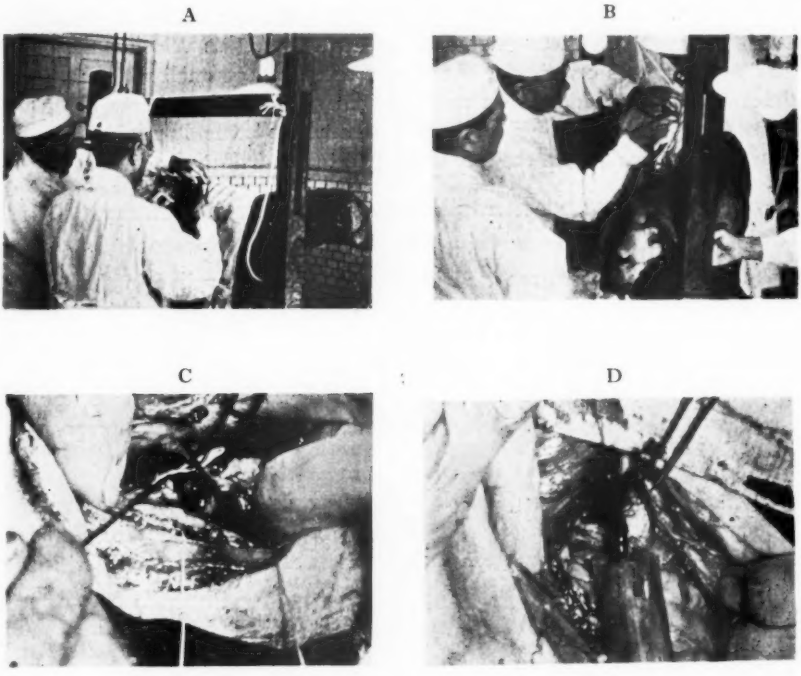


Fig. 1, A, B, C, D. Three-pieces from the lumbar route operation. The dog, the lumbar roots of which from the IX thoracic to the II lumbar were previously cut, is standing on the table. A: Fixing the sheet by stiching on the skin. B: Tying the cut end of a skin vessel after a vertical straight incision through the skin is now started closely to the last rib. C: The lateral surface of the gland is seen here laying nearly vertical and lumbo-adrenal vein crossing the gland; now a long, somewhat strong thread is pushed through behind the vein by means of a silver mounted bent needle. D: Blood is here running out from the cannula inserted and fixed in the adrenal vein just distal from the gland. (The procedure of the method is given in detail in Satake, Sugawara, and Watanabe, *Tohoku J. Exp. Med.*, 1927, **8**, pp. 503-507.)

The flow rate through the adrenal gland changes according to altered conditions; when an acceleration occurs in the secretion velocity, the blood flow also increases commonly, but it does so on a very small scale, i.e. three times *ad maximum*, in contrast to the enormous fluctuation observable in the increase of the adrenaline concentration, such as some hundred times *ad maximum*. During sleep the rate is reduced to one-half of the quiet rate.

(B) Adrenaline in the Venous Blood from the Adrenal Gland

In the experiments reported in this book the adrenaline in the venous blood from the adrenal gland was determined by means of the rabbit intestinal segment method,[5] and sometimes also by the method of the paradoxical pupil reaction of the cat in parallel with the former. Some remarks are given here in general in practising both the methods at first. Some different features between them are observable when applied to the rabbit during tis pregnancy.

Rabbit intestinal method:

This method was well developed by Stewart and Rogoff. When we proved the effectiveness of sensory stimulation upon the adrenaline secretion from the base of the cava pocket method experiments, we were strongly criticized by Stewart, who took it that we had not mastered the intestinal method.[6] However, that they were wholly mistaken in their judgement was proved in comparing the intestinal tracings originating from both laboratories, Cleveland and Sendai.[7]

The lowest limit assayable by the intestine strip, for example, is not a whit different in the curves from both laboratories. The fact that quite a different outcome was yielded between them is due in fact to the difference in the procedures in carrying out the cava pocket experiment; the use of anesthetics is here solely responsible for their failure to see an increase of adrenaline secretion on sensory stimulation. They frequently called their method "the intestinal method" in place of "the cava pocket method": their regard for their beautiful tracings is, highly probably, betrayed there. The intestinal strip method is not indispensable for supplementing the essential, that is to say, the principal part of the method. Provided narcotics, such as avertin, evipan, veronal etc. were applicable there, the Cleveland physiologists had already arrived at the correct conclusion.

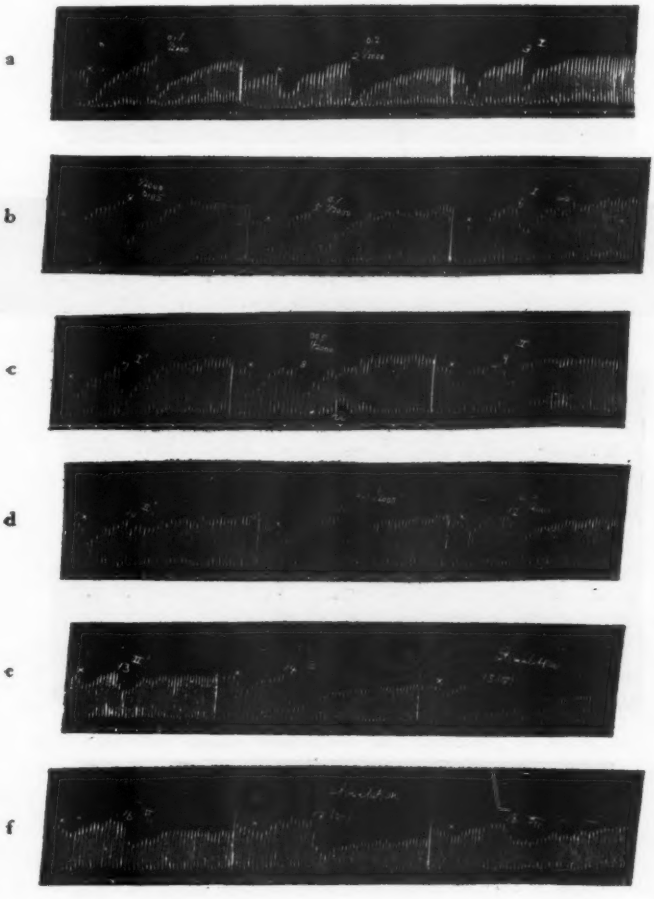
We could have immediately taken up the divergency of views about the intestinal method with the Cleveland physiologists at that time, but did not do so, because we did not wish to be polemic; alternatively, we repeated the experimentation anew, and it was almost two years later that we first answered.[7] At that time and afterwards we reproduced all intestinal tracings belonging to one experiment, without any omission. This shows how fairly

An Example of Intestinal Tracings

In all the intestine tracings, at the mark "X" atropine-Tyrode's solution, in which the rabbit intestine segment was beating rhythmically, was replaced by indifferrent blood solution, and at the "numeral" the indifferent blood solution to which a certain quantity of adrenaline chloride of Sankyo & Co. was added, or by the suprarenal vein blood solution. Quantity of all the bloods employed for one assay was 0.5 cc.

In all tracings, time intervals 30 seconds.

The number of the suprarenal vein blood specimen solution and the quantity of adrenaline hydrochloride in the adrenaline blood solution are written on every intestine tracing. The former with roman numerals; the latter in cc. of a given concentration. For example 0.2 1/2000 means 0.2 cc. of 1 :2,000,000 or 0.0001 mg.



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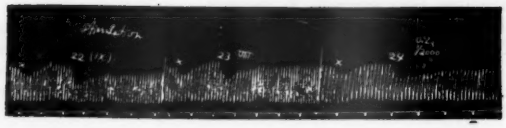

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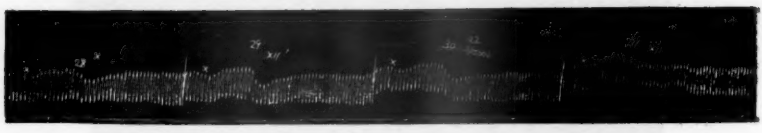

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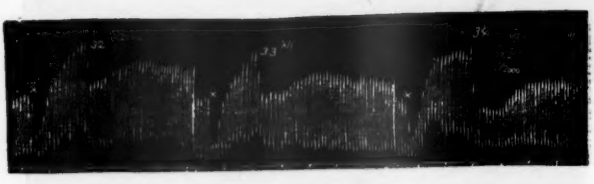

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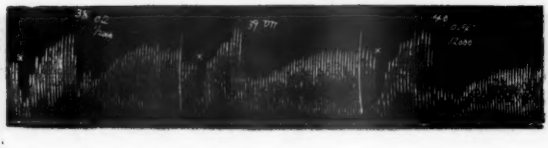

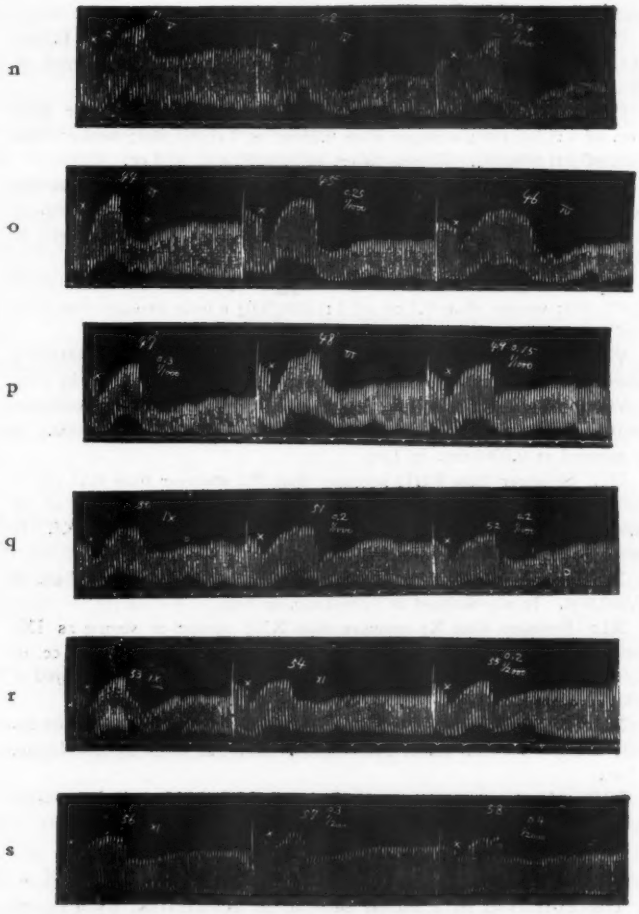

FIG. 2. An example of estimation of adrenaline in a set of experiments by means of the rabbit intestine segment. (Satake et al., *Tohoku J. Exp. Med.*, 1927, **9**, 27–31)

The results are to be summarized as follows:

I: Weaker than 0.2 cc. of 1:2,000,000; almost as strong as 0.05 cc. of 1:2,000,000; weaker than 0.1 cc. of 1:2,000,000; as strong as I′. I was assayed at 0.00005 mg. in 1 cc.

I′: Almost as strong as 0.05 cc. of 1:2,000,000; as strong as I. I′ was assayed at 0.00005 mg. in 1 cc.

II: Almost as strong as 0.05 cc. of 1:2,000,000; almost as strong as II′. II was assayed at 0.00005 mg. in 1 cc.

II′: Almost as strong as II; a little weaker than 0.1 cc. of 1:2,000,000;

weaker than III. II′ was assayed at 0.00005 mg. in 1 cc.

III: Stronger than II′; weaker than IV; almost as strong as 0.2 cc. of 1 :2,000,000; weaker than 0.3 cc. of 1 :2,000,000. III was assayed at 0.0002 mg. in 1 cc.

IV: Stronger than III; stronger than V (two assays); weaker than 0.4 cc. of 1 :1,000,000; stronger than 0.25 cc. of 1 :1,000,000; weaker than 0.3 cc. of 1 :1,000,000. IV was taken as 0.00055 mg. in 1 cc.

V: Weaker than IV (two assays); weaker than VI; almost as strong as or a little stronger than 0.3 cc. of 1 :2,000,000; stronger than 0.2 cc. of 1 :2,000,000; far weaker than 0.4 cc. of 1 :2,000,000. V was assayed at 0.0003 mg. in 1 cc.

VI: Stronger than V; stronger than VII; weaker than 0.25 cc. of 1 :1,000,000; weaker than 0.3 cc. of 1 :1,000,000; a little stronger than 0.15 cc. of 1 :1,000,000. IV was taken as 0.0004 mg. in 1 cc.

VII: Weaker than VI, almost as strong as 0.2 cc. of 1 :2,000,000; weaker than 0.4 cc. of 1 :2,000,000. It was assayed at 0.0002 mg. in 1 cc.

VIII: Weaker than IX; weaker than 0.2 cc. of 1 :2,000,000; stronger than 0.1 cc. of 1 :2,000,000; almost as strong as 0.15 cc. of 1 :2,000,000. It was assayed at 0.0001 mg. in 1 cc.

IX: Stronger than VIII; stronger than X; stronger than 0.15 cc. of 1 :1,000,000; almost as strong as or very little stronger than 0.2 cc. of 1 :1,000,000; almost as strong as 0.2 cc. of 1 :000,000; as strong as XI. It was assayed at 0.0004 mg. in 1 cc.

X: Weaker than XI; weaker than IX; almost as strong as 0.1 cc. of 1 :2,000,000. It was assayed at 0.0001 mg. in 1 cc.

XI: Stronger than X; stronger than XII; almost as strong as IX; stronger than 0.2 cc. of 1 :2,000,000 (two assays); stronger than 0.3 cc. of 1 :2,000,000; almost as strong as 0.4 cc. of 1 :2,000,000. It was assayed at 0.0004 mg. in 1 cc.

XII: Weaker than XI; weaker than 0.2 cc. of 1 :2,000,000 (two assays); stronger than 0.1 cc. of 1 :2,000,000. XII was taken as 0.00015 mg. in 1 cc.

XII′: Almost as strong as 0.2 cc. of 1 :2,000,000; a little stronger than X. It was assayed at 0.0002 mg. in 1 cc.

the method has been mastered there. We do not think much of a case in which the concentration in a blood sample is determined by a figure which is not reproduced there, while a large number of tracings are given there.

After several experiments we came to find an intestinal loop of about 15 cm. length extending above and below the entrance of the pancreatic duct of rabbits as a good test object—not only taken from a rabbit, which was killed in another experiment, but also from that on the previous day.[8]

Cat paradoxical pupil reaction:[9]

Small modifications, such as an injection of test fluids into the carotid artery,[10] and use of a reading microscope of magnification 9,[11] make somewhat sensitive the cat paradoxical pupil reaction, as described previously. Most

of the experiments given in this book however, were done commonly without these modifications.

By these modifications the paradoxical eye reaction has become satisfactorily available for estimation of a small amount of adrenaline in the blood, just as the rabbit intestine segent method.

In 40 dogs, in which our method (the lumbar route method in the deafferented dog) was applied and successive blood samples or one and the same blood were tested by the rabbit intestinal segment method and the cat paradoxical pupil reaction, the ratio of both methods was 1 : 0.7–1.5, mostly 0.8–1.1. Generally speaking they are almost equal.

(They are: 2 caffeine, 1 strychnine, 2 urethane and 2 tetrodotoxine by Aomura, 3 cold, 1 exercise and 6 cerebellum in Wada, and 2 acetylcholine, 9 nicotine, 4 lobeline, 5 pilocarpine and 3 physostigmine. There are no drugs which have an significant variation in this ratio.)

Thus the adrenaline content in the adrenal blood of dogs may be taken as identical, roughly speaking, when determined by means of the rabbit intestine segment or of the cat paradoxical pupil. A similar tendency is also visible in practising chemical methods such as those of Shaw or Bloor.

Rabbit blood collected for example by the cava pocket method, give definitely different figures, i.e. intestine: pupil 1 : 2 (1 : 1.2–1 : 4). Whole gland or medullary extracts of cattle (4.5% : 3%, 16.1% : 11.3%), pig (3.8% : 2.2%, 18.5% : 10.4%) horse (2.3% : 2.5%, 15.1% : 13.5%) and rabbit (whole gland, 0.5% : 0.6%) give the figures indicated in parentheses. With dogs we have no such experiences.

Variation of the content in the adrenal blood and gland of the adrenaline-intestine and adrenaline-pupil in the rabbit during pregnancy:[12,13]

Adrenaline preparation (of Sankyo) being taken as the standard, as usual, the cava pocket preparation shows: adrenal gland or blood commonly involves the adrenaline-pupil, which is determinable by the cat paradoxical pupil reaction, in about double the quantity compared with the adrenaline-intestine, determinable by the rabbit intestine segment. But when pregnant, their content varies clearly (See Fig. 3); the adrenaline-pupil decreases as the pregnancy advances and the adrenaline-intestine increases, so that on the 21st day of pregnancy, both the amounts become almost the same. On the 27th day the adrenaline-intestine becomes the greatest, and two days later both the amounts regain their original and normal values.

Fig. 4 shows the adrenaline content in the glands left behind after having conducted the cava pocket experiment shown in Fig. 3; it does not show peculiar figures, corresponding to those in Fig. 3, while Fig. 5 indicates almost similar figures as those of Fig. 3. This shows the adrenaline content in rabbits killed by a blow on the neck only on a certain day, of preg-

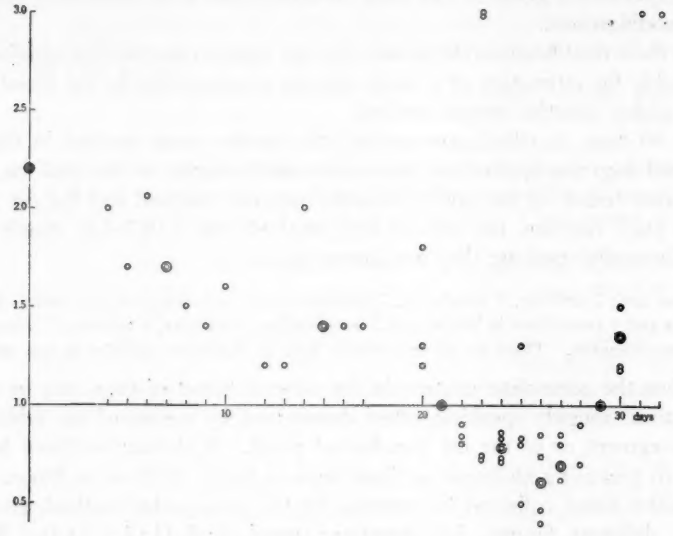

FIG. 3. Fluctuation of the ratio between the adrenaline-intestine and the adrenaline-pupil in the adrenal blood of rabbits in the course of pregnancy and a few days after birth. (Aomura)

In these three figures, light circles show the ratio of adrenaline-intestine to adrenaline-pupil in each test, and double circles the averages, taken during some days.

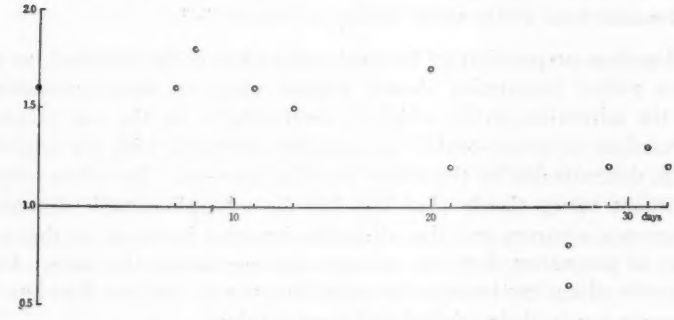

FIG. 4. Fluctuation of the ratio between the adrenaline-intestine and the adrenaline-pupil in the adrenal extract in the course of pregnancy and a few days after birth. (K. Saito)

nancy, without any other manipulation than that blow.[14] Tab. IV on p. 13 shows a similar tendency with these findings; a relation between the output

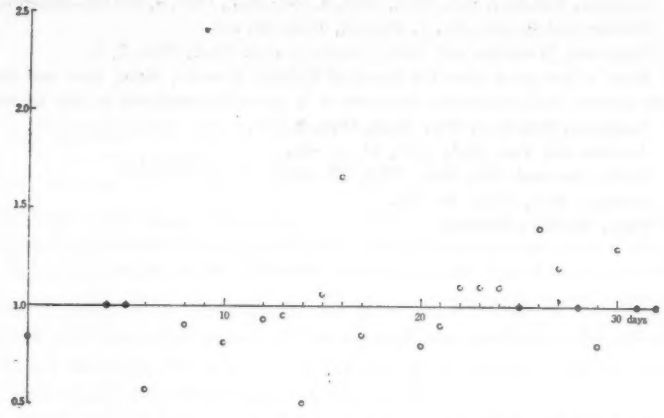

FIG. 5. The same as in Fig. 4., but in the adrenal gland left behind after having carried out the cava pocket experiment, the results of which are given in Fig. 3. (Aomura)

of adrenaline on sensory stimulation and that which remains thereafter are given there.

(C) NORMAL RATE OF ADRENALINE SECRETED FROM THE ADRENALS OF NORMAL DOGS

In the experiments on dogs, non-fastened, non-anesthetized and non-laparotomized, 268 in toto, the adrenaline amount was on an average in 0.084 γ in 1 cc. adrenal blood, mostly 0.05–0.1 γ in 1 cc.

In the left gland 0.83 $\gamma \pm 0.0030$ (201 cases), and in the right gland 0.09 $\gamma \pm 0.0057$ in 1 cc. (67 cases); in male animals (180 cases) 0.086 $\gamma \pm 0.0034$, and in female (88 cases) 0.082 $\gamma \pm 0.0043$.

The flow rate in one gland 0.1–0.7 cc. per kg.-min. and in both glands combined 0.6 cc. per kg. per min.

Secretion of adrenaline: From one gland 0.025 γ per kg.-min., from both glands 0.05 γ per kg.-min. (268 cases).

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7) Sugawara, Watanabe and Saito, *Tohoku J. Exp. Med.*, 1926, **7**, 1.
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CHAPTER VI

NERVOUS AND HUMORAL CONTROL OF ADRENALINE SECRETION

CONTENTS: The central mechanism, hypothalamus, medulla oblongata, cerebellum, thoracic cord—Pathway of adrenaline-secretory fibres through anterior roots of the spinal cord—Splanchnic nerves as the adrenaline secretory nerves—Nicotine, acetylcholine.

THE subject matters in this chapter are given in the order of the central mechanism of the adrenaline secretion, its subordinate portion in the spinal cord, the pathway through the anterior or ventral spinal routes, and the secretion of adrenaline after the denervation of the gland.

THE CENTRAL MECHANISM

In the central nervous system there are two places which have much concern with the adrenaline secretion: one in the hypothalamus and another in the medulla oblongata. There are abundant secretion of adrenaline by stimulation and abolishment of existing secretion by paralysis or non-effectiveness of reflex stimulation.

Hypothalamus.

To search for the central mechanism of adrenaline secretion in the hypothalamus is natural. Stimulation of a posterior portion there causes an adrenaline secretion.

Houssay and Molinelli, who made use of the Tournade and Chabrol method, stimulated the infundibular part of the third ventricle in the donor, and observed the blood pressure rise and the heart acceleration in the recipient.[1] Karplus and Kreidl saw a blood pressure elevation on a hypothalamus stimulation in a cat under curare, but after bilateral adrenalectomy the pressure effect still continued;[2] exact evaluation of these results are difficult. A similar outcome was obtained in cats by Beattie et al., with extrasystole as the indicator.[3] The bilateral adrenalectomy does not prevent the occurrence of extrasystole there. In cats, chloroform narcosis brings about extrasystole. Beattie et al. were able to interfere with its occurrence by removing the nerve supply to the heart along with that of the adrenals. Stimulation of the hypothalamus still causes the extrasystole even after the adrenals are removed. The posterior hypothalamic region is responsible in causing extrasystoles.[3]

Magoun, Ranson and Hetherington tried to stimulate the hypothalamus in cats under dial, the nictitating membrane, denervated acutely (for adrenaline)

or chronically (for sympathin) being taken as an indicator. The effective region distributes itself through the antero-posterior extent of the lateral hypothalamic area and the adjacent periformical region.[4]

Bender and Weinstein observed a delayed depressor and mydriatic effect (completely denervated pupil) in cats under nembutal by hypothalamic stimulation.[5]

In cats under chloralose, Brauner, Brücke, Kaindl and Neumayer stimulated electrically the hypothalamus, and the adrenal blood was determined of adrenaline with pilomotor action on isolated hairs of the tail of a cat and isolated uterus of a rat. Quiet secretion—adrenaline 0.032 γ. in 1 cc., noradrenaline 1.246 γ; on hypothalamic stimulation they increase to 0.160 γ and 3.738 γ.[6] Quite recently Suzuki and Arai collected blood samples by means of the cava pocket method in cats under evipan natrium narcosis.[7] Adrenaline was determined by the rabbit intestine, the Bloor and the Bullen methods. Stimulation of the ventro-medial or ventro-lateral portions of the hypothalamus secretion increases from 0.03–0.11 γ per kg.-min. to 0.5–5 γ. While Houssay et al. noted that the acceleration of adrenaline secretion is definitely greater, as five times as high, when the fourth ventricle is mechanically stimulated, than that of the hypothalamus, Beattie et al. are of opinion that the bulbar centre may be an important relay station in the projection pathway from the hypothalamus to the cord. The latter investigators cut the regio between the regio and thereafter, in other words, cut the hypothalamus between the anterior edge of the colliculi and the posterior edge of the pituitary fossa. Then chloroform brings about no more extrasystole.

Drugs which affect the cortico-diencephalic centres, such as morphia, camphor, caffeine, and guanidine affect the adrenaline secretion.[8]

Medulla oblongata.

Elliott[9] who carried out sensory stimulation in the cat under ether or decapitation and determined adrenaline in the gland by the blood pressure method, accepted existence of the mechanism controlling the adrenaline secretion in front of the corpora quadrigemina and behind the 2nd cervical cord. Tournade and others tried cocainization of the fourth ventricle floor in dogs, anesthetized and provided with the adrenalo-jugular anastomosis, and found no adrenaline running into the recipient dog.[10]

On the contrary, Cannon assumes the central mechanism for the adrenaline secretion to be in a narrow portion, such as immediately at the back of the corpora quadrigemina in the floor of the fourth ventricle (Exp. on March 31). A small acceleration, respectively retardation was noted in the denervated heart even after double adrenalectomy (Tabs. 2 & 3);[11] but this can be explained by the sympathin-liver.

On the other hand, piqûre diabétique produces an abundant secretion

of adrenaline. In the lumbar route preparation on the de-afferented dogs of Yen, Kaiwa and Wada, the hyperadrenalinemia was 0.2 γ to 1 γ per kg.-min. against 0.02 γ to 0.04 γ from both glands. The hypersecretion period covered two to three hours.[12]

The piqûre lesion was found 2 to 13 mms. above the calamus scriptorius and on the mid-line or a little deviated. Blood sugar at that time 0.16 to 0.2%. In four normal dogs the piqûre caused a hyperglycemia of 0.17% as the mean; no adrenaline experiment was run. The lesion was 3–6 mms. above the calamus scriptorius.[13] In two examples (Dogs 12 & 17)[12] the piqûre lesion was 13 or 9 mms. from the calamus scriptorius, and the maximum adrenaline secretion among this set of experiments was 0.5 γ per kg.-min. This corresponds with the outcome of Cannon who determined the central location of the piqûre in cats. It was 2 mms. behind the corpora quadrigemina. On the whole the nervous machinery of the piqûre could be taken as being located more widely in the floor of the fourth ventricle. Caudal portion may be a little weak.

Cerebellum.

A puncture on the lobulus posterior or medianus of the cerebellum produces in dogs an acceleration in the adrenaline secretion on a scale of about one-third of that yielded in the piqûre diabétique, the numeral data being given in Chapter VII.[14]

In a rabbit deprived of the superior cervical ganglion and instillated with atropine, Shimidzu[15] conducted a puncture on the cerebellum of Eckhard; the pupil dilated thereupon.

On the other hand Cannon and Rapport are of opinion that the cerebellum is clearly not essential to reflex secretion of adrenaline. In two cats, decerebrated, reflex stimulation resulted in accelerating the denervated heart after removal of the cerebellum.[11] This fact does not directly contradict the data above given.[14]

Further noteworthy is the fact that the blood sugar commonly does not increase. In nine cases,[13] only three are qualified with hyperglycemia, 0.17% in Exp. 4 & 0.12% in Exps. 5 & 9. The adrenaline secretion in these cases 0.2–0.1 γ adrenaline per kg.-min.; no difference compared with the rest.

Rogoff noted further an inhibitory centre of adrenaline secretion between the optic chiasma and the superior colliculi.[16]

Thoracic cord.

Once too much importance was laid by Stewart and Rogoff upon the thoracic cord as being the central machinery for the adrenaline secretion.[17-19] According to them the transsection of the spinal cord before the thoracic cord does not alter the "spontaneous liberation." Sometimes reduction occurs

there ; it is due, according to them, to a spinal shock. However " spontaneous secretion ", " ordinary output " or " normal output ", as sometimes related by them also, are under their experimental conditions. The liberation, such as 0.23 γ per kg.-min., is clearly too quick. Their experimental conditions, such as narcosis, fastening, laparotomy, etc. already interfere with the rate of adrenaline secretion. And in fact the transaction before the thoracic cord reduces largely the output of adrenaline in all the cases of dogs, and in half the cases of cats.

In general, stimulation of afferent nerves augments the secretion of adrenaline (A), and that of special nerves, such as of depressors, i.e. of the aortic nerve and of the carotid sinus nerves, reduces it (B).

(A) The fact that stimulation of afferent nerves augments in general the secretion of adrenaline is well known. The negative results of the Cleveland physiologists in this respect are due to their particular experimental conditions, especially the misuse of narcotics. That the experimentalists who followed the cava pocket method rather faithfully could still obtain several positive cases depended upon how deep their narcosis was. In the hands of Stewart and Rogoff the initial rate (ether or ether+morpnine) was 0.2 γ adrenaline per kg.-min., in Kodama (ether) 0.8 γ[20] and in Sugawara et al. (ether) 0.5 γ.[21] For the Cleveland physiologists to get a negative result in stimulation, and a small and narrow initial rate of output, indicates how strong the inhibitory influence was there. In cava pocket experiments without narcosis, one can invariably see a reflex acceleration of adrenaline secretion.[22,21] Such a difference is not due to different sensibility of the intestinal segment as they suggested, but wholly due to the depth of narcosis employed by them and by us.

Sensory stimulation was done usually for 20 to 50 seconds, mostly 30 seconds. The acceleration continued the stimulation by, say 10 seconds, sometimes more and sometimes almost none. Sensory stimulation is commonly accompanied by emotional disturbance. In an exceptional case[23] (Dog 7), which was fastened and operated on under non-anesthesia, the animal cried, struggled very furiously and uninterruptedly throughout fastening, 23 minutes after the route operation 3 γ per kg.-min. was estimated. Such a case is very seldom met with, however.

On the other hand, the initial, quiet output rate in the cava pocket method in non-anesthetized dogs is 0.7 γ per kg.-min. in Kodama and in Sugawara et al., that is a little smaller than the cava pocket method under narcosis of Kodama (0.8 γ), greater than Sugawara (0.5 γ) and distinctly greater than Stewart and Rogoff (0.2 γ). Such a difference is highly probably due to the total sum of the accelerating influence by sensory stimulation and the inhibiting effect of narcosis. The initial rate in the denverated cava pocket method is just ten times that in the denervated lumbar route preparation.

Influence of sensory stimulation upon the adrenaline output is almost the same in these three kinds of experiments (the cava pocket under narcosis, the de-afferented cava pocket and the de-afferented lumbar route). Only the values obtainable under narcosis are a little smaller; i.e. the inhibitory influence either on the pain partaking effect or adrenaline secreting mechanism, or both, act clearly. The acceleration due to sensory stimulation was 2.5 to 3 times in the denervated lumbar route method of Satake et al. (6 cases), and 2 times in narcotized cava pocket of Kodama (12 cases) and of Sugawara and others (8 cases). In general the effect of sensory stimulation respectively of narcosis on the secretion of adrenaline depends upon the pre-existing rate of secretion.

Commonly the reflex acceleration of adrenaline secretion is rather small in comparison with other expressions against the sensory reaction; at least, that is our impression. Incidentally, the effect or ability of an agent upon the adrenaline secretion may be taken as an indication of how heavily the basal rate of secretion will be multiplied thereby, but its effect upon the whole organism must be considered first on the base of the absolute quantity of adrenaline secretion increased there.

Existence of a central mechanism of adrenaline secretion in the thoracic cord is evidenced in the experiments of the Cleveland physiologists.[24] Their spontaneous liberation takes place even after the last cervical cord section. The reduction occurs there, but how much the spinal shock acts there, is difficult to decide.

The rate of adrenaline secretion after the last cervical cord section in dogs and cats is about one-half to one-seventh of the initial.

Many agents affect the adrenaline secretory machinery in the thoracic cord, but some agents do not. For example, strychnine,[24] sensory stimulation,[25] peptone,[25] bleeding,[25] etc. act on the thoracic cord, but insulin does not directly act there.[11] It is however difficult to decide how the former agents act on the higher central machinery.

The ratio of increase in the adrenaline secretion in bleeding, peptone, and sensory stimulation to the initial rate is almost the same in the thoracic dog as in the normal dog, similarly prepared with the cava pocket preparation under narcosis. The doses of bleeding and peptone must be taken into account, of course; and the lethal effect of a large dose to an animal seriously operated on is also obviously to be borne in mind.

In the cases of sensory stimulation there were many negative results. As opposed to these cases, Stewart and Rogoff obtained in cats good evidence of the accelerating influence of strychnine upon the adrenaline in the cava pocket experiments in the cat under ether and sectioned under the cervical cord.

Contrary to the above agents, there is an agent which does not act upon

the thoracic machinery of adrenaline secretion, but only on its upper central part, as in the case of the insulin. For example, when insulin given in an i.v. dose of 7 to 8 units per kg. to such a dog, no acceleration takes place in the secretion rate of adrenaline. Hypoglycemia in such a dog is rather remarkable there;[26] it was 0.01 to 0.05% in these dogs (4 examples) in contrast to those of Yen and others[27] (3 cases) with the intact spinal cord, the values being 0.04 to 0.06%.[26]

Further, it may be added here that Jourdan reported the adrenaline secretion centre to be in the cervical cord.[28] In a dog under chloralose, the cervical cord was cut between the first and second. The cervical cord was circulated with the carotid and jugularis, the vagi and stellate ganglia being interfered with. If the vessels be closed the heart accelerated. On stopping the entrance of adrenal blood into the general circulation, it does not occur. The work does not seem to be elaborate.

(B) Agents which cause reflex inhibition in the adrenaline secretion are stimulation of the central end of the vagus[29] and of the depressor nerves,[30] i.e. of the aortic nerve and the carotid sinus nerve.

Pathway of Adrenaline-secretory Fibres through Anterior Roots of the Spinal Cord

Adrenaline-secretory fibres from the centre in the medulla oblongata or higher region of the central nervous system start in each half of the medulla oblongata and divide in the thorcic cord bilaterally, then through the anterior roots and finally through the splanchnic nerves to the adrenals.

Originating from the adrenaline-secreting centres, which are situated according to Chen, Lim et al. near the inferior fovea, presumably quite the same location as the vasomotor centres, the fibres reach the ventrolateral column through the unilateral pathway.[31] After hemisection on the cervical cord 7. to 8., insulin can still produce an adrenaline secretion from both adrenal glands.[32] Suzuki et al. further saw no adrenaline secretion due to insulin from the ipsilateral side when the cervical cord was cut through between C_7 and C_8 on one side, and the thoracic cord between both sides. After the hemisection at the thoracic cord D_2 a stimulation on the spinal cord thereabove causes an adrenaline secretion from both adrenals.[33]

There are then some experiences obtained by Kodama and Sugawara from 1921,[31] on dogs with dissection of the dorsal roots from the fourth thoracic cord to the second lumbar cord. The cava pocket experiment was carried out with the basal rate of adrenaline secretion of 0.66 γ per kg.-min. No narcosis was used. This is almost in the same range as the cava pocket experiment on normal dogs under anesthesia. Sensory stimulation increases the adrenaline secretion and ether narcosis reduces it. In the lumbar route preparation, carried out thereafter in this laboratory for some decades, the

dorsal roots from the ninth thoracic to the third lumbar cord were cut off beforehand. Similar data have been yielded there, as this monography shows.

Some similar figures were yielded in the hands of Algerian physiologists.[35] In their adreno-jugular anastomosis experiments of dogs, stimulation of the ventral roots in quite the same scope or limits accelerated the adrenaline discharge, and centrigugal stimulation of the dorsal roots of the thoraco-lumbar cord had no effect at all.

However, since there is a school persisting the authenticity of the passage of the adrenaline-secreting fibres in the dorsal roots of the spinal cord,[36] Masao Wada[37] in our laboratory sectioned on the one side the dorsal roots from the fourth or fifth thoracic cord to the third or fourth lumbar cord, and on the other in the ventral side two or three upper roots in the thoracic cord and three to four lower roots in the lumbar cord besides those mentioned above. The additonal sections were done to obviate pain at the lumbar route operation.

Morphine was then intravenously given in a dose of 8 mg. per kg., resulting in a clear demonstration of the data wholly harmonizing with our expectation.[38] In the cases of the dorsal root section, the adrenaline secretion occurred in quite the same manner as in the normal animal, and none at all in those of the ventral root section. Morphine acts to liberate adrenaline by the central action only. Further, that the peripheral mechanism was wholly intact in the last cases was proved by the use of nicotine.

Kuré school's atropine experiments which support a view that parasympathetic fibres in the splanchnic nerves convey only the impulse for the secretion of adrenaline, and the sympathetic only that for its production are shown to be wholly untenable.[39] The contrary evidence is cogent enough; so there should never be the least hesitation in giving up the contention.

Splanchnic Nerves as the Adrenaline Secretory Nerves

Cutting the splanchnic nerves, major and minor, in advance or just beforehand, reduces largely the secretion of adrenaline from the adrenal gland. There are a few papers reporting its total abolishment (Stewart and Rogoff, and Sugawara on nicotine). Further, some lumbar sympathetic and coeliac ganglia were taken out besides splanchnicotomies. It is not yet sure whether one is able to abolish totally the secretion of adrenaline thereby.

Basal rate of adrenaline secretion from the adrenal gland of dogs is 0.025 γ per kg.-min. (About three hundred experiments. Rabbit intestinal method), and 0.013 γ per kg.-min. after cutting the splanchnics major and minor. From both glands the basic rate is 0.05 γ per kg.-min., and after splanchnicotomy 0.025 γ per kg.-min.[40]

While thus the splanchnicotomy diminishes the basal rate of adrenaline

secretion by a half in the non-narcosis lumbar route preparation, it acts so by one-tenth in the narcosis cava pocket experiments.

In the ether cava pocket experiments of Kodama, Sugawara and Watanabe, the basal rate of adrenaline discharge after splanchnicotomy was 0.06 γ per kg.-min., while that with the intact innervation in Kodama and Sugawara was 0.05 γ–0.08 γ per kg.-min. In the narcosis cava pocket experiments of the Cleveland physiologists, the former was 0.005–0.01 γ.[41] This is also about one-tenth of the basal rate of those with intactly innervated adrenals, roughly speaking. The adrenaline secretion lasts under experimental conditions such as those of Tournade after the splanchnicotomy major et minor as in the mass of 0.005–0.007 γ per kg.-min.[42]

The secretion from a denervated gland is sometimes called the sécrétion paralytique. However, it is very difficult to use the phenomenon observed by Cl. Bernard as an indicator of adrenaline secretion, as elsewhere related. From the present technique we see quite different figures from those reported by the French master of physiology.[43]

The vagus has no direct connection with the adrenals.[44]

The adrenaline secretory agencies are divided into two groups, those which can not be activated at all after excluding the splanchnic nerves and the others still working.

Group A. Adrenaline secretion of a central nature, i.e. no secretion of adrenaline, if applied after cutting off the major and minor splanchnic nerves.

Such as asphyxia,[45] strychnine,[46] β-tetranaphthylamine hydrochloride,[47] caffeine, diuretine,[48] morphine,[49] insulin,[50] peptone,[51] histamine.[52] Of the effect of sensory stimulation this action is parenthetically related to.

Group B. Some substances cause adrenaline secretion from the denervated adrenal gland. Nicotine, its allied substances, acethylcholine, pilocarpine, physostigmine, urethane, potassium chloride, etc.

Nicotine:

Nicotine accelerates enormously the adrenaline secretion first, then depresses it for a long time.[52–55] The enormous acceleration is a salient feature of action, and the depressant period is also a typical phenomenon there.

Cutting of the major and minor splanchnic nerves scarcely altered the adrenaline secretion due to nicotine at all, or showed some reduction. When nicotine in a dose of 1 mg. i.v. was given by Wada et al., adrenaline was estimated ad maximum as 10 γ–0.3 γ adrenaline per kg.-min. in dogs with normally innervated adrenals, and 4.5–2.4 γ adrenaline per kg.-min. after denervation. In the experiments of Watanabe, who started to collect a little later after the injection, an intravenous injection of 1–0.75 mg. nicotine per kg. caused an oversecretion of adrenaline such as 0.1–0.3 γ adrenaline per kg.-min. while the glands were innervated, and 0.06–0.2 γ adrenaline per kg.-min. after

denervation. The variation is due solely to that of the adrenaline concentration.

The above data come very close to the findings of S. Kojima in cats, revealing that the threshold dosis of nicotine for the paradoxical pupil dilatation does not alter by splanchnicotomy.[54]

In ether cava pocket experiments the accelerated adrenaline secretion due to nicotine was diminished by one-fifth after double splanchnicotomy.[55] The basal rate and the accelerating action of nicotine run parallel before and after the operation.

That nicotine acts hyperadrenalinemic by a periphery action has been reported from various laboratories. In these cases a problem still remains as to whether it has also a central action.

Nicotine acts on the adrenal medulla itself or further on chromaffine cells in general. The postganglionary machinery of the sympathetic nervous system is fluid there in the adrenal medulla.

Some writers have reported that cytisine, lobeline, tetramethylammonium, conicine, etc. are similar in the adrenaline secretory action with nicotine.[56]

Acetylcholine:

Acetylcholine can produce adrenaline secretion from the adrenals even when the central nervous system or the splanchnic nerves with the upper abdominal sympathetic chain be excluded.[57] The question still remains whether acetylcholine acts so also on the central nervous system, and whether it acts with stronger effect when the adrenals are denervated.

Quantitative data show that acetylcholine induces an abundant secretion of adrenaline even after double splanchnicotomy.

In dogs with the lumbar route preparation under non-anesthesia E. Inaba injected acetylcholine subcutaneously in a dose of 15 mg. per kg.[58] After the denervation a rather stronger secretion was obtainable, 0.4 γ adrenaline (0.7–0.2 γ) per kg.-min. in 3 dogs against 0.1 γ (0.07–0.2 γ) adrenaline in 5 normal dogs. This is due to the change of adrenaline content.

A later series of similar experiments in which the drug was intravenously injected in a dose of 3–5 mg. per kg. in 30 seconds, showed the highest speed as 0.5 to 1.5 γ per kg.-min. in 4 intactly innervated dogs and 0.6 to 2 γ per kg.-min. in 4 doubly splanchnictomized.[59]

Therefore it is sure that acetylcholine causes the adrenaline secretion abundantly without intervening with the splanchnic nerves.

That the minimal effective dose of acetylcholine for the paradoxical pupil reaction is reduced in cats on double splanchnicotomy harmonizes with these events. In this experiment neither anesthesia, nor fastening was utilized.[60] The main event here cited corresponds well with the fact that acetylcholine can reduce the adrenaline content in rabbits, irrespective of whether the

splanchnici are retained intact or previously cut off.[61]

Potassium chloride:

A generally accepted fact is that potassium chloride acts to accelerate the adrenaline secretion, partly centrally and also partly peripherally on the adrenal medulla.[62] Its injection in the fourth ventricle of the brain causes adrenaline secretion.[63]

Pilocarpine,[64] physostigmine,[65] urethane[66] also induce adrenaline secretion after splanchnicotomy. Drugs which are proved to accelerate adrenaline discharge from the denervated organ, can act so by a direct injection into an adrenal gland or on perfusing it.

Finally it might be added here again that adrenaline does not act to accelerate the adrenaline secretion. The general arterial and venous blood samples must be collected with the adrenal blood simultaneously at such an experimentation.[67]

How the dorsal root section acts is a question which must be taken into account in experiments with such a lumbar route preparation. We have not yet found any appreciable data here. For example, the quiet rate of adrenal blood flow in cava pocket experiments under ether was 1 cc. (0.8–1.8 cc.) per kg.-min. in 17 dogs with normal roots, and 1.5 cc. (1.3 cc.–1.8 cc.) in 5 deafferented dogs.[71] Here only some references are to be added to the report of 1924.[72]

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CHAPTER VII

SECRETION OF ADRENALINE FROM THE ADRENAL GLANDS

CONTENTS: A. General, B. Particular

A. General*

CONTENTS: Variation of the rate of adrenaline secretion from the adrenals depends solely or chiefly upon that of the adrenaline concentration of the adrenal blood—Acceleration of the adrenaline secretion—Depression of adrenaline secretion.

CERTAIN agents effect an increase in the rate of secretion of adrenaline from the adrenals, some others a decrease therein, and the third group of agencies no effect.

A great number of materials, especially those used in medicine might be probably included in the third group. However, as a matter of course, the materials without action upon the adrenaline secretion have been examined or reported rather seldom.

The subject, here tested by means of the lumbar route preparation on the non-anesthetized, non-fastened dog, the operation field of which was beforehand de-afferented, are as follows: Asphyxia (for 1.5–9 minutes), CO (3 or 5%, 20 litres, inhalation in 2 minutes), cyanide (2–2.5 mg. per kg., i.v.), low pressure (200 mm. Hg.), hemorrhage (1/3–2/5 of the total blood volume), peptone (0.1–0.3 g. per kg., i.v.), histamine (1–4 mg. per kg., i.v.), anaphylactic shock, insulin (3–10 units per kg., i.v.), guanidine (0.15–0.3 g., i.v.), sensory stimulation (20–30–60 seconds), piqûre diabétique, cerebellar puncture, muscular exercise (fatigue), cooling, heating, anesthetics and hypnotics (ether, chloroform, chloralhydrate, chloralose, urethane (1–3 g., i.v.; 1 g., subc.), veronal, evipan, avertin), adrenaline (0.05–0.3 mg. per kg., i.v.), acetylcholine (5 mg. per kg., i.v.; 15 mg. per kg. subc.), pilocarpine (1–4 mg., subc.), physostigmine (0.5 mg., subc.), atropine (5–10 mg. per kg., i.v.), nicotine (1 mg. per kg., i.v.), morphine (8 mg. per kg., i.v.), strychnine (0.015–0.1 mg. per kg., i.v.), caffeine (0.15 g. per kg., i.v.), etc., ephedrine (0.03 mg. per kg., i.v.), lobeline (0.03 mg. per kg., i.v.), camphor (2.5 & 5 g. per kg. subc.), vitacamphor (i. v.), digitalis (i.v.), rhodein (i.v.), tetrahydro-betanaphthylamine (1–5 mg. per kg., i.v.), sodium nitrite (20–30 mg. per kg., i.v.), potassium chloride (20–30 mg. per kg., i.v.) and tetradotoxin (0.3–4 mg. per kg., subc.).

(The figures in parentheses indicate the doses mostly applied, and the mode of application.)

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* Reference to literature on the subject, quoted in the general introduction of Chapter VII is in most cases given in the respective place where discussed. There are available, however, a number of reports relevant to the last three paragraphs of this chapter, besides those quoted in this book. Reference to the reviews in the books of Trendelenburg, Bayer, Hartman, etc. recommended.

Variation of the Rate of Adrenaline Secretion from the Adrenals Depends Solely or Chiefly upon that of the Adrenaline Concentration of the Adrenal Blood

Adrenaline is secreted from the adrenal glands with a fairly constant rate, if reckoned on the animal's body weight, so long as the animal behaves normally. This comes in reality from the data that both the flow rate of the adrenal vein blood and the concentration therein are constant. In dogs, generally speaking the rates are 0.05 γ per kg.-min., 0.6 cc. per kg.-min., and 0.085 γ per cc. respectively, as shown in Chapter V.

The variation, increase or decrease, in the rate of adrenaline secretion, is solely or chiefly effected by the concentration rate of adrenaline in the adrenal blood.

The blood flow through the adrenals at most doubles in the experiments with insulin, piqûre, morphine, and acetylcholine on the one hand, and decreases at most by one-half in those with hemorrhage, peptone, anaphylactic shock, cold bath, etc. on the other hand.

The scale of the variation, either increase or decrease in the blood flow from the adrenals, is however extremely small in comparison with that of the adrenaline concentration therein. And only in the cases where the secretion rate varies in a small measure, has the variation in the flow rate any bearing thereupon. Chloralose, avertin, veronal are such examples. In many other cases the blood flow fluctuates only insignificantly; sometimes it increases, sometimes decreases, and sometimes shows practically no change at all.

From this data it follows that in future it will be in order to measure the variation in the adrenaline secretion rate by inserting the opening of a catheter into the adrenal vein, with a minimum of disturbance to the organism itself —apart from that caused by the insertion—and by estimating adrenaline in the blood samples collected through the catheter. Non-estimation of the blood flow out of the adrenals does not materially impede the determination of the secretion rate variation. To ascertain the secretion rate itself, the rate of adrenal blood flow must, needless to say, be measured too.

Acceleration of the Adrenaline Secretion

CONTENTS: Rate—Total amount of oversecretion—Duration of oversecretion—When the oversecretion reaches its peak—Types of the course of oversecretion of adrenaline—Relation of exitement or depression in animals to the occurrence of hyperadrenalinemia—Hyperadrenalinemia and the splanchnic nerves—Whether an adrenalinemic agent acts on the thoracic cord or only on the higher mechanism—Analysis of action upon the central mechanism of adrenaline secretion.

As to the acceleration of adrenaline secretion brought about by the various agents given above, the data appertaining to the degree of acceleration,

its duration, the total amount of oversecretion, etc. will now be set forth.

Of each agent, various doses were tried, and in most instances increased up to the lethal quantity, as by so doing it was possible to determine with certainty its greatest hyperadrenalinemic effect.

The numerical figures in this chapter applying to the secretion rate of adrenaline, the adrenaline concentration, the duration of oversecretion, the total amount of oversecretion, etc. show the average only, if not otherwise noted. For example, the highest rate of adrenaline by nicotine is on an average 8 γ per kg.-min., as given below, while 20 γ was in fact the highest among all nicotine experiments.

Rate.

The most way to express the degree of acceleration in the adrenaline secretion is by the ratio of the accelerated rate to the preliminary rate as elsewhere discussed. Since the latter is remarkably constant (0.05 γ adrenaline per kg.-min. from both glands), it is also satisfactory and convenient to express it by the absolute value.

Roughly speaking, rates of from 0.1 γ to 10 γ per kg.-min., i.e. accelerations of 2 to 200 times were measurable in dogs under the influence of various agencies mentioned.

The accelerating agencies are here arranged according to the hyperadrenalinemic power: the classification is schematic, and some subjects are included in successive groups.

TABLE VI

The Maximum Rate of Adrenaline Secretion Caused by Various Agents

10 γ adrenaline per kg.-min.	two hundred times: Nicotine
5 γ ,, ,, ,,	one hundred times; CO (inhal.), KCl, furious excitement (1 example)
3 γ–1 γ ,, ,, ,,	twenty to sixty times: potassium cyanide, acetylcholine, asphyxia, caffeine, hemorrhage, anaphylactic shock, peptone, histamine, piqûre diabétique, low atmospheric pressure
0.5 γ ,, ,, ,,	ten times: morphine, hemorrhage, peptone, histamine, insulin, guanidine, piqûe diabétique, ephedrine, camphor (subc.), digitalis, rhodein
0.5 γ–0.2γ ,, ,, ,,	five times: sensory stimulation, cerebullar puncture, cooling, heating, urethane (i.v.), pilocarpine (subc.)
0.1 γ ,, ,, ,,	two times: muscular exercise (exhaustion), physostigmine (subc.), atropine, strychnine, lobeline (subc.), vitacamphor, β-tetra, $NaNO_2$, cold water into stomach, urethane (subc.), tetradotoxin (subc.)

For reference the increase in the adrenaline secretion on splanchnic stimulation is quoted here from the cava pocket experiments of the Cleveland physiologists.[1] The greatest rate in 7 dogs under ether was 3 γ adrenaline per kg.-min.

The value actually obtained is given here doubled, because the major splanchnic nerve

was stimulated there on one side. The authors noted this, whereby 10 times acceleration was calculated there, but if the rate of 3 γ per kg.-min. were found under our experimental conditions, it would be a 50 times acceleration.

Total Amount of Oversecretion.

For evaluating the adrenalinemic ability of various agents it is a matter of great importance to elucidate the total amount of oversecretion of adrenaline. It is not reasonable to arrive at the acceleration's multiplier by comparing the highest rate with the preliminary one.

For example, potassium, chloride, on the one hand, is capable of causing as great a rate as 5 γ adrenaline per kg.-min., but the total amount of oversecretion is so small as to be less than 0.05 γ per kg., while the piqûre diabétique, on the other hand, brings about 0.5 γ and 100 γ respectively.

Again, to arrive at the total amount of oversecretion of adrenaline it is not sufficient, as a matter of course, to note the highest rate and the length of time of acceleration. One must precisely follow the rate of secretion along the whole course of secretion.

The total amount of oversecretion by various agencies is averaged in each, and then schematically arranged according to degrees:

TABLE VII

Total Amount of Oversecretion by Various Agencies

100–50 γ adrenaline per kg.	: hemorrhage, piqûre diabétique
75 γ ,, ,, ,,	: insulin
50 γ ,, ,, ,,	: anaphylactic shock, guanidine, caffeine, ephedrine, camphor (subc.), digitalis, rhodein
25 γ ,, ,, ,,	: CO (inhal.), histamine, morphine, nicotine (somewhat less), urethane (i.v.)
10 γ ,, ,, ,,	: asphyxia, peptone, cerebellar puncture, cooling (somewhat large), urethane (subc.) sodium nitrite (somewhat large)
5 γ ,, ,, ,,	: cold water into stomach (somewhat small), heating, physostigmine (subc.), strychnine, tetradotoxin (subc.)
3–2 γ ,, ,, ,,	: potassium cyanide, acetylcholine, pilocarpine (subc.), vitacamphor, β-tetra
Less than 1 γ ,, ,, ,,	: KCL, lobeline (subc.), muscular exercise (exhaution)

Oversecretion exceeding 100 γ per kg. was discovered in some cases of peptone, piqûre diabétique, hemorrhage. In pilocarpine poisoning a large oversecretion was exceptionally estimated (20 γ per kg.).

It should be noted here that both adrenals together of dogs contain, roughly speeking, 200 γ adrenaline per kg. as the mean value estimable by the chemical method of Suto-Kojima or of Folin, Cannon and Denis, and 300 γ by the rabbit intestine segment method.

The adrenalinemic action of each agency is commonly expressed by comparing the maximum rate with the basal, as above; it is simple, but

by no means sufficient even insomuch as to indicate the grade of ability. The total amount of oversecretion can be taken into account in doing so, and it is further of significance to compare the total amount of oversecretion with the amount of adrenaline, which would be secreted provided the animal were at rest during the period as long as the oversecretion exceeds the effective limit. When applied uniformly for two minutes, 0.2 γ per kg.-min. adrenaline is given as the least dose for increasing the denervated heart rate or the arterial blood pressure in dogs, otherwise quite normal.[2] For the present 0.1 γ is taken here as the limit value, because the oversecretion of adrenaline nearly always rapidly advances, and this rate was noted as being such limit value in some experiments of adrenaline infusion.

Accelerating action of various agencies are appoximately calculated in this manner, and given here according to the grades.

TABLE VIII

Grades of Acceleration of Adrenaline

(The total amount of effective oversecretion : the basal secretion in the corresponding period)

15 times : asphyxia, carbonmonoxide, nicotine, acetylcholine
10 times : hemorrhage, anaphylactic shock, peptone, histamine, ephedrine, camphor, potassium chloride, potassium cyanide
5 times : piqûre diabétique, insulin, guanidine, digitalis, morphine, cooling in bath
3 times : urethane, cerebellar puncture, cold water into stomach, heating, vitacamphor
2 times : strychnine, physostigmine, etc.
Only small : muscular exercise (exhaustion), pilocarpine, lobeline

Duration of Oversecretion.

This varies largely according to the different agents; it varies from a minute to three hours or longer. The figures that follow actually show what a significant bearing this factor has in making the total quantity of adrenaline secretion, and in determining the grades of acceleration shown in the above tables.

TABLE IX

Duration of Adrenaline Secretion by Various Agents

3 hours : hemorrhage, insulin, guanidine, piqûre diabétique, urethane
2 hours : peptone, histamine, anaphylactic shock, cerebellar puncture, pilocarpine, physostigmine (subc.), morphine, strychnine, ephedrine, camphor (subc.), digitalis, sodium nitrite, tetradotoxin (subc.)
1 hour : muscular exercise (exhaustion), cooling in bath, cold water into stomach, heating, atropine, caffeine, lobeline (subc.), β-tetra
30 minutes : CO (inhal.), vitacamphor (i.v.), nicotine
10 minutes : asphyxia
3–2 minutes or less : potassium cyanide, acetylcholine, potassium chloride, sensory stimulation

When the Oversecretion Reaches its Peak.

There exists some relation between the duration of oversecretion and the time when its peak appears, but it is dissimilar depending upon the agents or groups of agents. For instance, the ratio of the latter to the former is 1 : 25 for peptone, histamine, anaphylactic shock, and morphine, and 1 : 3 for hemorrhage and insulin. The relationship plays a significant part in determining the type of the course of adrenaline oversecretion. Tab. X shows when the peak takes place.

TABLE X

When the Oversecretion of Adrenaline Reaches its Peak

1 hour: insulin, pilocarpine (subc.), physostigmine (i.v.), strychnine (90 minutes), ephedrine, camphor (subc.), rhodein (?), sodium nitrite

30 minutes: hemorrhage, piqûre diabétique, guanidine (20—), cold bath, β-tetra

20 minutes: cerebellar puncture, cold water into stomach, heating, lobeline (subc.), digitalis, tetradotoxin (sub., 10 minutes)

5 minutes: peptone, histamine, anaphylactic shock, acetylcholine (subc.), morphine, caffeine, vita-camphor (i.v.)

2–1 minutes: asphyxia, potassium cyanide

Under 1 minute: acetylcholine (i.v.), nicotine (15 seconds), potassium cyanide (15 seconds), sensory stimulation

Whether a drug be applied intravenously or hypodermically has a significant bearing upon the time when the acme of oversecretion appears, and the whole duration of oversecretion is a fact readily comprehensive, and indicted in the above tables too.

Long latencies were observable frequently in the experiments with urethane (i.v., subc., 30 minutes or longer), pilocarpine (subc., 20 minutes or longer), physostigmine (subc., 15 minutes), lobeline (subc. 10 minutes), digitalis, rhodein (both, i.v., 5 minutes), and insulin (i.v., 10–15 minutes). That the insulin adrenalinemia has a long latency is to be expected if the mechanism of producing it be taken into consideration.

In passing it should be pointed out that if application of agencies be followed immediately by the collection of adrenal blood samples, and subsequent samples be successively taken with only short intervals of time between them, the latency could still be detected with accuracy. But even allowing this, no latency was detected in the experiments in which adrenaline was secreted extraordinarily rapidly, as in poisoning with nicotine, acetylcholine, carbonmonoxide. This does mean however that the latency of adrenaline secretion is shorter than the period of the first sample collection in our experiments, i.e. thirty seconds or the like.

Types of the Course of Oversecretion of Adrenaline.

Generally the adrenaline oversecretion has only a peak; otherwise ex-

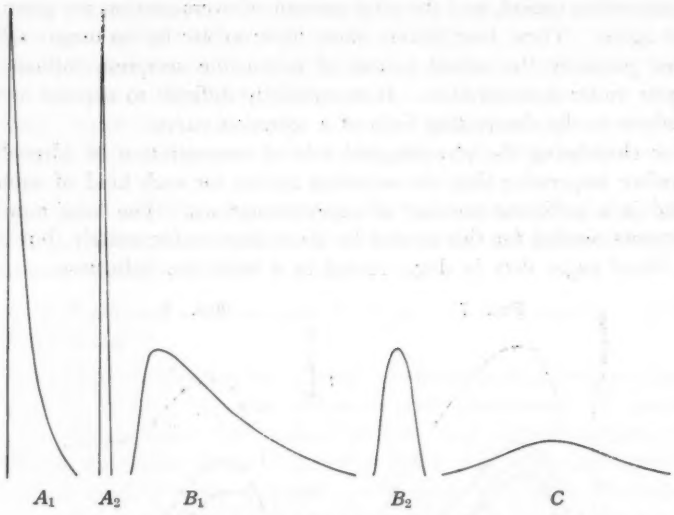

FIG. 6. Types of oversecretion of adrenaline from the adrenal gland of the dog, schematically drawn.

pressed, an increasing and a decreasing period.

In some cases the velocity with which the secretion increases and that with which it decreases are almost the same, but in many other cases the reduction progresses distinctly slower. The latter may be called Type 1 because of high frequency of occurrence.

On the other hand the rate of oversecretion varies largely according to various kinds of agencies, as is amply proved here, so that secretion courses or curves can be classified according to the greatness of secretion rate, schematically into three classes of speediness, great, intermediate and small. To differentiate in this way is merely a matter of expediency, of course.

With both factors combined we get Type A_1, Type A_2, Type B_1, Type B_2, and C. There are a great number of oversecretions of Type B_1, and only a few cases of C type.

A_1-type: CO (inhal.), KCN, nicotine, caffeine
A_2-type: KCL, acetylcholine (i.v., subc.) (More difficult to decide of KCl, ACH.)
B_1-type: asphyxia, hemorrhage, peptone, histamine, anaphylactic shock, insulin, guanidine, piqûre diabétique, cold bath, heating, nicotine, morphine, digitalis
B_2-type: camphor (subc.), vitacamphor
C-type: urethane (i.v., subc.), physostigmine (subc.), strychnine
The oversecretion curve due to hemorrhage has double peaks

The rate of the greatest speed, the time when it is arrived at, the length

of oversecretion period, and the total amount of oversecretion are given above of each agent. These four factors alone however are by no means sufficient to show precisely the actual course of adrenaline secretion influenced by the agent under consideration. It is especially difficult to express or depict fluctuations in the descending limb of a secretion curve.

For elucidating the physiological role of oversecretion of adrenaline, it is therefore imperative that the secretion course for each kind of subject be followed in a sufficient number of experimentations. The least number of experiments needed for this cannot be given here unfortunately, but on this point, blood sugar data in dogs, cooled in a bath, are indicative.

FIG. 7 FIG. 8

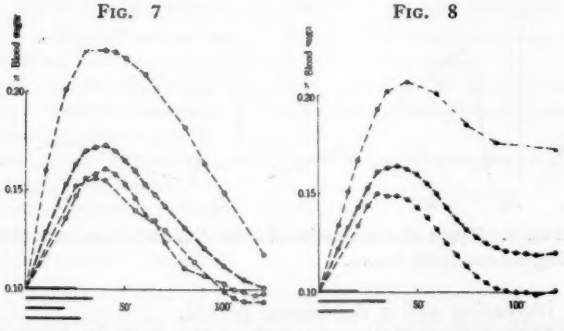

FIG. 7. Blood sugar fluctuation in normal dogs on cooling in the cold bath. Three sets experiments: From top to bottom, 4 dogs (Satow), 6 dogs (Itikawa) & 4 dogs (Wada et al.). Thick broken line shows the average of all the cases (14 dogs). The thick solid, straight lines on and under the abscissa show the average time of cooling in each set.

FIG. 8. Blood sugar fluctuation in medulli-adrenalectomized dogs on cooling in the cold bath. From top to bottom 4 dogs (Satow), the mean (15 dogs in toto), and 11 dogs (Itikawa). Further see the legend for Fig. 7.

The hyperglycemic curve was uneven in three sets of experiments (4, 6 & 4 dogs) in normal dogs, and in two sets (11 & 4 dogs) in medulli-adrenalectomized animals, but if the sets are grouped to give 14 dogs in the one group and 15 in the other, completely smooth curves are obtainable. The curve composed from 11 experiments, although almost smooth, is not so smooth as the last mentioned curves. And as to findings applicable to the adrenaline secretion, ten experiments are needed at least to fix a general figure of hyperadrenalinemia; fifteen will give more satisfactory data.

Adrenalinemic curves in peptone and anaphylactic shock were, roughly speaking, more or less smooth; they are composed from 6 experiments on each; and in fact the conditions causing the shock were not particularly uniform. Peptone was dosaged from 0.05 g. per kg. to 0.3 g., or the horse

serum was intravenously introduced in various amounts until the blood pressure began to fall. (See Fig. 14 on p. *23 L*)

If a desire to have in hands the general pattern for each agent, not only in form, but also in the quantitative sense, could be realized, it would be very much appreciated. It would greatly facilitate study on the physiological significance of the secretion of adrenaline and other sympathomimetic hormones.

The adrenal medulla is by no means the " rois fainéant " of the endocrine glands.[3] Our data, given in this book, clearly show, and quantitatively verify, the existence of the medulla's function.

Relation of Excitement or Depression in Animals to the Occurrence of Hyperadrenalinemia.

The method of collecting the adrenal vein blood in dogs without fastening, narcotizing or evoking any pain affords a good background for gaining an insight into the problem the heading shows. Any indirect evidence, which is obtainable by means of the denervated heart, e.g., while the state of the animal is not ideal, is to be evaluated just as such, only when it corroborates the results of a more direct method such as this.

With this direct method we saw in a strong dog a furious emotional excitement accompanied by an enormous hyperadrenalinemia, viz. 6γ adrenaline per kg.-min., i.e. an acceleration of one hundred times.

Reflex stimulation brings about violent, aggressive reaction in normal dogs on the one hand, and 2–5 times, at most 8 times acceleration in the secretion rate, roughly 0.1–0.5γ adrenaline per kg.-minute, on the other hand. Similar violent struggles are elicited for a while by some strongly adrenalinemic agents as: asphyxiation (rate: 50 times acceleration, 3γ per kg.-min.; total amount 10γ per kg.), hemorrhage (10 times, 0.5γ, 60γ), cooling in bath (5 times, 0.25γ, 10γ), caffeine (40 times, 2γ, 40γ), etc.

Sensory stimulation and hemorrhage were here proved as still capable of causing oversecretion after excluding the central secretory mechanism above the thoracic cord. While it is small in cases with reflex stimulation, hemorrhage induces an oversecretion not very inferior to that in the control, if the condition of the animal be taken into consideration.

There are on the other hand some subjects which bring about a copious oversecretion, but only slight and transitory excitement. Camphor, piqûre of Bernard, peptone shock are examples (All, 20–60 times, 1–3γ & 50γ).

Taking collectively all the data here given, it might be unreasonable to deny any role played by emotional disturbances in causing the oversecretion where such disturbances take place, but it is very difficult to evaluate their role. In the second group of agencies, however not much weight should be attached to the emotion.

Narcosis depresses both the phenomena, excitement and oversecretion, hand in hand.

Lastly there are some subjects which promote the development of profound shock. Their application is immediately followed by convulsions, which not long afterwards are replaced by collapse; peptone, insulin, and the like. They cause also an abundant outflow of adrenaline (Peptone, etc. 20 times, 1 γ per kg.-min. & 25 γ in toto; insulin, etc. 10 times, 0.5 γ & 75 γ). And the interrelationship between the depression and the oversecretion is quite intimate. So long as the depression lasts, the oversecretion is maintained: they run parallel completely.

Hyperadrenalinemia and Splanchnic Nerves.

The basal rate of adrenaline secretion is reduced by half on sectioning the splanchnic nerves. General figures are 0.05 γ adrenaline per kg.-min. and 0.025 γ respectively. When sleeping it is 0.02 γ per kg.-min. (Cf. this chapter, Particular, p. 80)

A majority of hyperadrenalinemic agencies acts through the splanchnic nerves to the adrenal medulla; a few others do not.

Oversecretion of adrenaline, causable by the following agencies was proved here, i.e. with dogs, unfastened, unanesthetized, as entirely or almost entirely depending upon the intergrity of the innervation: asphyxia, CO, peptone, histamine, insulin, morphine, strychine, caffeine, etc. Splanchnicotomy largely reduces oversecretion of adrenaline brought on by pilocarpine and potassium chloride too.

Nicotine and acetylcholine cause an enormous discharge of adrenaline from the adrenal gland, deprived of its innervation. Giving 1 mg. nicotine per kg. produced oversecretion of 8 γ adrenaline from both glands per kg.-min. and 20 γ adrenaline in toto, while they were not yet denervated, and that of 7 γ per kg.-min. and 16 γ per kg., after the denervation was carried out. With 5 mg. acetylcholine per kg. an oversecretion of 2.4 γ kg.-min. & 2 γ per kg. in toto, and 2.1 γ per kg.-min. & 1.3 γ per kg. in toto respectively resulted.

It might possibly be justifiable to assume from these figures the identity of the intensities of oversecretion due to both drugs, before and after the denervation of adrenals.

Despite the vivid central action of nicotine in general, no similar evidence has been detected hitherto for the adrenaline liberation from the adrenals. (Further, Cf. Chapter VI, p. 42-3)

Whether an Adrenalinemic Agent Acts on the Thoracic Cord or only on the Higher Mechanism.

Almost all agencies, other than those that act on the adrenal medulla

itself, act, to be sure, on the central mechanism for regulating the adrenaline secretion, but what portion or portions and to what extent each of those agencies directly or indirectly acts to effect the secretion has yet to be determined.

From a technical point of view it is however by no means difficult to decide whether an agent acts on the nervous mechanism in the thoracic cord or in the higher portion. However, only a few subjects have been tested in regard to this query: strychnine,[1] asphyxia and reflex stimulation are capable of causing oversecretion in the thoracic cord dog, while insulin acts only on the central mechanism situated thereabove.

Analysis of Action upon the Central Mechanism of Adrenaline Secretion.

A large number of subjects acting centrically, result in the adrenaline secretion being accelerated or depressed. Some adrenalinemic agencies suggest by their respective characteristics the manner in which they stimulate the central apparatus of secretion. Asphyxia, carbonmonoxide, potassium cyanide, a markedly low atmospheric pressure and bleeding produce oversecretion of adrenaline most likely by abnormally low oxygen pressure in the central apparatus or by hindering the oxidative process there. Insulin causing a diminution of the blood sugar also in the central mechanism acts to increase the adrenaline secretion; the onset of the oversecretion, however, does not occur so early as when other subjects are administered, and so long as the blood sugar content diminishes beyond a defined limit.

No doubt is left of existence of emotional oversecretion of adrenaline. The oversecretion by peptone and the like is accompanied by a rapid fall in the blood pressure, but on the other hand associated invariably with the depressive state of the animal. Whether both events are causally related with the oversecretion must be left as open to question.

Depression of Adrenaline Secretion

The depression may be produced by narcosis generally; this results chiefly or solely by a reduction in the concentration of adrenaline in the adrenal vein blood.

The depressive action is clearly demonstrable in the following manner: a dog, the operation area of which was beforehand de-afferented, is fastened prone on a table and the cava pocket is then prepared, without narcotizing. The preliminary rate is now, say, 0.65 γ adrenaline per kg.-min., then ether is given, which results in a reduction in the secretion rate of adrenaline to 0.25 γ per kg.-min. Alternatively, the cava pocket is prepared in a dog, narcotized with ether; cava pocket samples taken, say, ten minutes after stopping the etherization, and a rate of 1.2 γ adrenaline per kg.-min. is ob-

tained. On giving then ether, it is now estimated as 0.5 γ. The examples show that etherization reduces the liberation by 0.4 γ and 0.7 γ per kg.-min. respectively, or both by 60%. The coincidence shows, it might be noticed, that the deepness of anesthesia is in both experiments the same.

Generally, the deeper the anesthesia, the greater the depression in the secretion rate of adrenaline.

The rate of the so-called spontaneous liberation of adrenaline, measurable in the cava pocket experiment in dogs under ether was 0.2 γ per kg.-min in the hands of Stewart and Rogoff,[5] 0.8 γ in Kodama,[6] and 0.5 γ in Sugawara.[7] The figures indicate the different depths of anesthesia. The Cleveland physiologists gave too much anesthetics, and in consequence, were unable to find any accelerating action of reflex stimulation, asphyxia, etc. upon the adrenals' secretion.

P. Trendelenburg, who properly evaluated how far our method could go in building up or elaborating the physiology of adrenal medulla, failed however to indicate any recognition of a definite depressive action of narcosis thereupon, when referring to Kodama's paper.[8] This however was not unreasonable at that time: in fact such a view was then a general trend. Glycemia and adrenalinemia develop hand in hand in most cases, and ether glycemia is a well established fact, and so on.

Depressive ability of narcosis can be expressed only by a reduction in the scale of increase in the adrenaline secretion causable by given agencies or under a given condition or conditions. The degree of depression is thus expressed by the ratio of the absolute rate or amount of reduction to the rate or amount prevailing before that depression. See the examples given ahead in this paragraph.

Generally surgical anesthesia, as ether or chloroform act strongly or even finally nullify the adrenaline secretion due to asphyxia, sensory stimulation, hemorrhage, anaphylactic shock, the like, insulin, piqûre, etc., while urethane, on the contrary, alone cause an oversecretion of adrenaline.

On the other hand there are some agencies, the adrenalinemic ability of which is not strongly influenced by surgical anesthesia. They are pilocarpine, strychnine, morphine, etc. Atropine and β-tetranaphthylamine hydrochloride produce an almost similar degree of oversecretion, irrespective of whether anesthesia is resorted to or not. Only larger amounts of the agents are necessaty there for producing the oversecretion.

Aliphatic hypnotics act to reduce the oversecretion of adrenaline, but only on a very small scale, or almost nothing at all. Chloralhydrate, chloralose, evipan and veronal were tested as such by us here.

(Further see Paragraph " Narcotics " on pp. 94–98.)

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1) Stewart and Rogoff, *Am. J. Physiol.*, 1924, **69**, 605.
2) Wada and Z. Kanowoka, *Tohoku J. Exp. Med.*, 1936, **27**, 9.
3) Cf. Stewart, *Arch. Int. Med.*, 1929, **43**, 764.
4) Stewart and Rogoff, *J. Exp. Med.*, 1917, **26**, 613; *Am. J. Physiol.*, 1920, **51**, 484.
5) *Idd., Am. J. Physiol.*, 1923, **66**, 235; *Ibid.*, 1924, **69**, 605, etc.
6) Kodama, *Tohoku J. Exp. Med.*, 1924, **4**, 601.
7) Sugawara, Watanabe and S. Saito, *Ibid.*, 1926, **7**, p. 73 ff.
8) Trendelenburg, *Die Hormone*, Berlin 1929, 313 & 326.

B. Particular

CONTENTS: Asphyxia—Carbonmonoxide—Potassium cyanide—Hemorrhage—Peptone—Histamine—Anaphylactic shock—Carotid sinus nerve—Insulin—Guanidine—Sensory stimulation, Fastening, Emotional excitement, Sleep—Piqûre diabétique—Cerebellar puncture—Muscular work (Exhaustion)—Cold application—Heat application—Narcotics and Hypnotics (Ether, Chloroform, Chloralhydrate, Chloralose, Urethane, Alcohol, Veronal, Evipan, Avertin)—Adrenaline—Acetylcholine—Pilocarpine—Physostigmine—Atropine—Nicotine—Morphine—Strychnine—Caffeine, Diuretine, Theophyline—Ephedrine—Lobeline—Camphor, Vitacamphor—Digitalis—Rhodein—Tetrahydro-betanaphthylamine—Ammonium chloride—Magnesium sulphate—Natrium nitrite—Potassium chloride—Diphtheria toxin—Tetanus toxin—Tetradotoxin.

This section of the chapter is chiefly composed of data from the experiments in which a given stimulation is applied to the dog, which has been prepared for collecting the adrenal venous blood in quiet, almost normal, or physiological conditions, as in the manner described in Chapter V. The adrenaline was estimated by means of the rabbit intestine segment method. In some instances the cat paradoxical pupil method was used in addition.

Remark 1. The rate of secretion of adrenaline given in this monography, i.e. in texts and figures as well, is generally the value from both adrenals, while the gland on one side is commonly experimented on in "the lumbar method."

Figures in this book which show curves of the adrenaline secretion, blood sugar fluctuation, etc. are plotted against the time indicated on the abscissa. Of estimates given on the ordinate, zero in the adrenaline secretion, 0.1% in the blood sugar concentration, 39°C. in the anal temperature, the value at the start of measurement in the arterial blood pressure and that in the pulse rate are plotted on the origin. The former three values expressed are actual ones, and the latter two are in the percentage of actual value against that at the start, this being taken as 100. The agency was commonly given or started at the origin (e.g. injected).

In general the scales of the values and time are the same throughout all the figures; only in a few cases where the secretion of adrenaline takes place at an extraordinarily enormous rate or when it fluctuates very rapidly, are their scales suitably adjusted. Such are cases with nicotine, carbon monoxide, cooling, etc., and the adjustment is especially annoted in the legend under the figure.

By way of precaution it should be pointed out that the rate of secretion, blood sugar, pulse rate, etc. here commonly reproduced in the figures with constant scales might erroneously lead the reader to conclude that there is some relationship in respect to the greatness of variations of the factors. The reason for such an arrangement, however, is that it has the merit of making it easy to compare the variations of all factors in several figures.

In most of the cases the blood sugar, arterial blood pressure, heart rate and anal temperature were simultaneously determined. In certain cases the denervated heart reaction or the paradoxical pupil reaction was also determined at the same time with the collection of adrenal venous blood. The outcome re the blood sugar, denervated heart, and paradoxical pupil will be particularly dealt with in Chapter VIII with those of the adrenaline secretion, and discussed there in advance of their interrelationship.

General remarks on the figures given in Chapters VII and VIII.

In all figures each subject is indicated respectively with a special form of line, as given below.

Solid line ——	adrenaline, rate of secretion in γ per kg.-min.; pupil width in Figs. 26 & 27.
Dash line — —	blood sugar, in per cent.
Dotted line ·····	blood pressure, in mm. Hg.
Chain line •–•–	heart rate, in beats per min.
Double line =	body temperature, in °C.

On the original are marked: 0 of adrenaline secretion rate, 0.1% of the blood sugar content, 100 mm. Hg. of the arterial blood pressure, 100 beats per min. of the pulse and 39°C. of the anal temperature. The scale of each subject and the time is quite the same throughout all the figures, but there are a few exceptional cases. This makes the data in almost all the figures readily comparable with each other. The exceptions are of such cases where the adrenaline secretion reaches an extraordinarily great rate or varies quickly; they are Figs. 9, 32, 35 A & B in part, 36, 37, special remarks are noted there. Time scale is modified there accordingly.

Experiments were carried out on dogs under various conditions, i.e. on the normal dog, the dog with bilateral medulli-adrenalectomy, or with double splanchnicotomy, or the dog with the completely denervated heart or pupil or further with their combined operations. In some cases an intravenous infusion of adrenaline solution in imitating the augmented secretion in the control dog or the like was simultaneously run.

The figures obtained under these various conditions are indicated with the following symbols:

Remark 2. The pulse rate given in the figures in this book are not the actual rates, but those recalculated on the basis of the body temeprature. The rate of the excised heart varies according to the temperature of the heart itself. The heart rate, recalculated in this manner, when compared with the rate at the start of experimentation, enables one to quantitatively estimate the influence of a certain factor or factors other than the temperature, such as adrenaline, sympathin, etc. (Cf. Chapter VIII)

Remark 3. Literature which is directly or indirectly related to the problems presented in this chapter, has been already as exhaustively as possible referred to in our previous papers. In this chapter, however, those which have direct or indirect but intimate bearings on our quantitative data, or the ideas deduced therefrom, are as a matter of course, quoted here again, and evaluated and discussed anew.

Remark 4. Of the pulse rate, blood pressure, and anal temperature the original protocols have been consulted, because they were not fully reproduced in the previous papers. The pulse rate has been recalculated on the above given principle; the method is given in Chapter VIII.

Light circle ○	The normal dog, given with a stimulus
Solid circle ●	The dog with double medulli-adrenalectomy, given with a stimulus; the rabbits or dogs (in Figs. 12 & 22) with double adrenalectomy, given with a stimulus
Light circle with point ⊙	The same, with the adrenaline infusion
Cross ×	The same, with a stimulus given and the adrenaline infusion; the adrenalectomized dogs treated similarly to the former (Fig. 12)
Double quadrate ▣	The same, with a stimulus given and the saline solution infusion; the adrenalectomized dogs, similarly treated (Fig. 12)
Light triangle △	The dog with double splanchnicotomy with a stimulus given
Solid triangle ▲	The same, with the adrenaline infusion
Double triangle ◬	The same, with a stimulus given and the adrenaline infusion
Light quadrate □	The dog with the denervated heart, with a stimulus given
Solid quadrate ■	The dog with the denervated heart and double medulli-adrenalectomy, with a stimulus given

These measures together facilitate the comparison of the figures with each other.

The data given in this following paragraphs are roughly in the order of agents producing anoxemia, blood pressure fall, hypoglycemia, some effects on the central nervous system or the body temperature, narcotics, autonomic hormones and poisons, inorganic salts, bacterial toxins, and so on.

Asphyxia

For asphyxiating the normal dog, certain manipulations other than the asphyxiation itself are necessary.

For example: the dog is first muzzled, though not tightly, lifted by the head and legs, and gently placed on its side; the mouth and nose are then wrapped around with a folded wet towel, having a thin rubber plate between the folds. The dog is asphyxiated in this manner. When the respiration becomes laboured and slow or wholly stopped, a matter of from 1.5 to 9 minutes, the wet towel is taken off and the animal set free; the animal then usually begins to breath spontaneously: if it does not, artificial respiration has to be conducted.[1]

Asphyxiation produces in the normal dog an oversecretion of adrenaline (7 experiments). The peak of oversecretion appears in a few minutes, schmatically 1–2 minutes following the asphyxiating period; then the rate goes back rather rapidly to the initial. The longest duration of oversecretion was a half hour. The mean of the greatest rate was 0.8 γ, per kg.-min.; the highest 3 γ. Ten minutes after having reached the peak the initial rate was almost recovered; 20–30 minutes after discontinuation of asphyxiation the rate was completely normal. The course of oversecretion of adrenaline might thus be assumed as of Type A_2.

The total oversecretion of adrenaline due to the asphyxiation in such an amount as to stop the respiration and render the dog unconscious, is

computed at 2–4 γ usually, or on an average 3 γ per kg., the greatest being 10 γ per kg.

The oversecretion is due chiefly at least to an increase of adrenaline concentration in the blood. It depends wholly upon the integrity of the splanchnic nerves (3 experiments).

The maximum rate of adrenaline secretion on asphyxiating the dog under ether or chloralose is 7 γ per kg.-min., as reported by Kodama,[2] and 2 γ by Houssay.[3] Such experimental conditions as those are sufficient to cause an oversecretion of adrenaline *per se*, as discussed elsewhere in this book.

In the experiments on the foregoing normal dogs, the secretion was not estimated during the asphyxiation. The cava pocket experiment on anesthetized dogs will therefore now be quoted to supplement the lacunae there.[2] The oversecretion already starts within twenty seconds of asphyxiation and advances as the asphyxiation is maintained for a further thirty seconds. The secretion taking place immediately after the end of asphyxiation of sixty seconds might be of a still greater rate.

A few words may be added here in conclusion: it has been well established that the asphyxia, especially if deep, is capable of eliciting glycemic, pressoric, and paradoxical pupil reaction, though less, even after excluding the adrenal secretion from the circulation or cutting the splanchnic nerves.[2,4] According to Stewart and Rogoff the denervated heart may be accelerated in cats in the amount, not less than normal.[5]

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2) Kodama, *Ibid.*, 1923, **4**, 166.
3) Houssay and Molinelli, *Revista Soc. Argent. Biol.*, 1925, **1**, No. 5; *Am. J. Physiol.*, 1926, **76**, 533.*
4) Itikawa, *Tohoku J. Exp. Med.*, 1936, **28**, 7ff.
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CARBONMONOXIDE

The method applied for letting the normal dog, provided with the "lumbar route" preparation, inhale CO-air mixture was as follows: a cannula with inspiratory and expiratory valves was inserted into the trachea under local anesthesia. Then 3% CO-air mixture was released for inhalation therethrough, e.g. 20 litres[1] in 2 minutes. About twenty to thirty minutes later 5%–3% CO-air mixture was further given in an amount of 10–16 litres.[1]

In the first test the blood pressure and pulse rate mostly increased, while

* When one and the same investigation was published in two or more periodicals, only the paper more common and readily available is given in this book, provided it does not interfere with the priority, so far as the references here are concerned.

on the second inhalation the reverse invariably resulted. In both the tests the pulmonary ventilation increased largely and the breathing rate increased. While the blood pressure rose, the animal struggled strongly.

Carbonmonoxide inhalation induces an enormously rapid but transitory increase in the rate of secretion of adrenaline, of Type A_2, as indicated in Fig. 9. Simultaneously the arterial pressure either ascends or falls as explained above.

Taking all the cases together, the mean maximum rate is 3γ per kg.-min., the greatest one being 10γ. That is: CO inhalation can induce usually some hundred times acceleration of adrenaline secretion, and a maximum of 1 000 times. The acceleration is due almost entirely to an increase of the concentration, and depends solely on the integrity of the splanchnic nerves.

The latency of oversecretion i.e. the time elapsed from the start of inhalation to the beginning of the secretion rate increase was roughly speaking one minute, and the time when the peak appeared was on an average two

FIG. 9

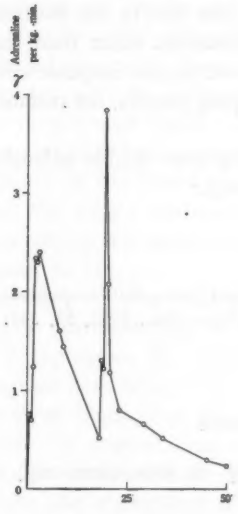

FIG. 10

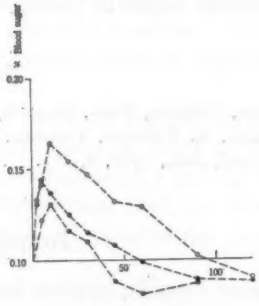

FIG. 9. CO-inhalation twice (Wada et al.); adrenaline secretion soon starts to increase, the latency being measured as 1 min.. Adrenaline scale is here taken as 1 : 10 of the common in this book, and time scale as 2 times.

FIG. 10. Blood sugar fluctuation on CO-inhalation (Suzuki, Takahasi & Tamabuti). From top to bottom: CO to normal dogs, CO to medulli-adrenalectomized, and adrenaline hydrochloride saline solution to the medulli-adrenalectomized.

to three minutes after beginning the inhalation. Duration of the inhalation was about two minutes or so.

The hyperadrenalinemic period covered half an hour to one hour, roughly speaking. The total amount of adrenaline, extraordinarily secreted on carbonmonoxide inhalation, conducted in the manner above noted, was 20-30 γ per kg. body weight.[1]

Inhalation of 3% CO in a total amount of 2 500–8 000 cc. causes in normal dogs a hyperglycemia with the mean acme of 0.18–0.19%, and an excess of sugar of 3 g. in 60–90 minutes. The demedullation of adrenals reduces it to 0.14%, and 1.1 g. in 60 minutes.[2] An infusion of adrenaline hydrochloride saline solution in imitation of the natural adrenaline secretion in CO-inhalation in amount and time course, brings about a hyperglycemia with 0.13% peak, 0.6 g. excess amount and 30 minutes duration. (Cf. Fig. 10) Inhalation of the gas in a large amount such as 20 000 cc. of 3% CO is capable of producing a large hyperglycemia both in the normal dog and in one without the adreanl medulla (0.21%, 4 g. and 0.28%, 4 g.).

These figures show, broadly considered, that the blood sugar increase due to CO can be attributed to two factors, one due to the oversecretion of adrenaline, and another to the glycemic mechanism other than the adrenal medulla. Both mechanisms are set into action by the impulse sent through the splanchnic nerves. Only a very small hyperglycemia, for instance 0.11%, is detectable after double splanchnicotomy.[1]

The hyperglycemia due to CO, remaining after double splanchnicotomy seems somewhat larger in rabbits than in dogs.[3]

References

1) Wada, Kitahara, Tano, Magoshi, Onodera and Saito, Will be published.
2) Suzuki, K. Takahasi, Tamabuti *Tohoku J. Exp. Med.*, 1952, **55**, 347.
3) Mikami, *Ibid.*, 1926, **9**, 113.

Potassium Cyanide

Potassium cyanide, applied intravenously or subcutaneously, causes an oversecretion of adrenaline.

On i.v. injection of 2–2.5 mg. per kg., the secretion is augmented to the rate of 4 γ per kg.-min., and to the total amount of 3 γ per kg. The maximum rate is measured one minute after injection, and the whole period of oversecretion one and a half minutes.

The blood sugar increases; the arterial pressure elevates for a while on i.v. injection, is soon replaced by a large fall, while subcutaneous injection causes no definite, significant variations.

REFERENCE

Tat. Suzuki, K. Saito and Kobayasi, *Tohoku J. Exp. Med.*, 1951, **54**, 343.

LOW ATMOSPHERIC PRESSURE

A low pressure, 198 mm. Hg., corresponding to 10 000 meters height, invariably results in an increase of the adrenaline secretion rate and the blood sugar concentration. The maximum was 0.9 γ per kg.-min. and 0.15% respectively. The O_2 and CO_2 content in the chamber was 22% and 0.4%.

A pressure of 267 mm. Hg., corresponding to 8 000 meters height, reached in 40 minutes, is not sufficiently low to bring on such changes.

REFERENCE

Wada, H. Sato, S. Satoo, Tano, Moriya and Semba, Will be published.

HEMORRHAGIA

Hemorrhage of certain intensities invariably produces in the normal dog an oversecretion of adrenaline from the adrenals.[1,2]

The blood was taken from a femoral or lumbar artery. Usually a few minutes to ten minutes or longer was needed for letting out one-third or a little more of the total blood quantity.

The minimum effective amount for effecting an oversecretion of adrenaline is one-fifteenth of the total blood quantity, and the greater the amount the greater and the longer the oversecretion, generally speaking. One-third or two-fifths might be the maximum effective amount, since too large an amount is detrimental to life.

On bleeding of one-third of the total quantity the dog responds with an adrenaline secretion rate of ten to thirty times as quick as the initial; numerically expressed, the maximum rate is mean 0.5 γ per kg.-min., the actually highest case being 0.8 γ. The oversecretion lasts for, schematically expressed, three hours, and the total amount of oversecretion is mean 60 γ per kg., with the biggest case of 100 γ.[1,2]

Adrenaline oversecretion associated with hemorrhage is characterized commonly by the appearance of two peaks, as Fig. 11 indicates. The first peak appears half an hour after bleeding, and the second two hours or more after bleeding.

The blood sugar increases with the peak, detectable ten minutes or later, so that it precedes the first peak of adrenaline secretion. No second elevation occurs in the blood sugar curve.

Narcosis reduces the effects of hemorrhage upon the adrenaline secretion. In the cava pocket experiment on the dog under ether, hemorrhage of one-

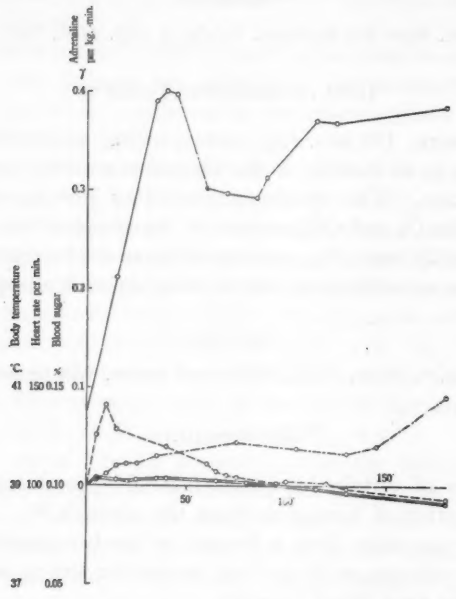

FIG. 11. One-third of the total blood shed in normal dogs; adrenaline secretion, blood sugar, heart rate & anal temperature are estimated. (Saito, Kamei & Tachi)

fifth of the total blood amount can cause a small but definite acceleration of adrenaline secretion. The lumbar route experiment, carried out under ether yields a similar outcome.[1]

In the dog, the spinal cord of which was divided between the cervical and thoracic cord, hemorrhage causes an oversecretion of adrenaline of a somewhat smaller degree. About half the rate seen in the normal dog is observable there: the effect is somewhat more pronounced when another stimulant, sensory stimulation for example, has been applied some time before. If the behaviour of the animal after having the cord sectioned before the thoracic be taken into consideration, we see indications that the hemorrhage definitely acts on the central mechanism in the thoracic cord, whereby the adrenaline secretion, above numerically given, can be occasioned.[3]

Data on the blood sugar will be referred to here so far as they have some bearing upon the adrenaline oversecretion.[4]

One third hemorrhage induces a large hyperglycemia, such as on an average 0.2%, 3 hours duration and 8 mg. excess sugar in toto; the largest cases 0.3%, 3 hours and 20 mg., and the smallest 0.13%, 2 hours and 2 mg.

The glycemic impulse is for the most part sent through the splanchnic nerves. The cutting of nerves reduces the glycemia to 0.12% on an average.[4]

On the other hand, the removal of the adrenal glands has little effect upon the hemorrhage hyperglycemia, but the clinical symptoms are more severe; the arterial pressure falls largely, and many of our animals died. As to the outcome in hyperglycemia in the adrenalectomized dog, one will remember that the same figure was commonly obtainable in the hands of Stewart and Rogoff in adrenalectomized rabbits; this will be fully discussed in the next chapter.

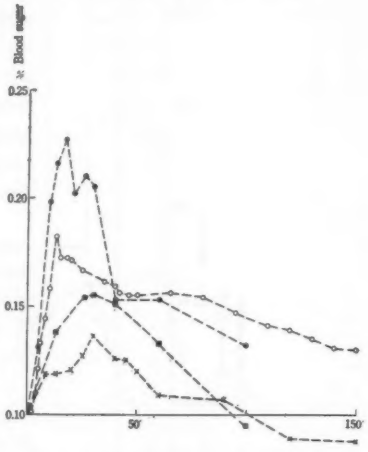

FIG. 12. Blood sugar fluctuation on bleeding of one-third of the total blood volume. (Sato, Kaiwa & Wada)

In normal dogs (light circles), in medulli-adrenalectomized dogs (solid circles), and the same in the dogs so operated on with simultaneous i.v. infusion of saline solution (double quadrates) or of adrenaline hydrochloride-saline solution (crosses).

It is further noteworthy that adrenaline infusion done in imitating the adrenaline oversecretion due to hemorrhage acts, on the contrary, to reduce the increase of blood sugar which should be brought about by bleeding, when the two—the infusion and the bleeding—are conducted simultaneously.[5] Introduction of saline solution serves as the control to the adrenaline infusion. The latter solution contains adrenaline hydrocholride of Sankyo Co., the saline solution as the vehicle. Adrenaline hydrochloride, intravenously given in the same manner, brings about a sufficient increase in the blood sugar, when applied to a doubly splanchnicotomized dog. (Fig. 13).[6]

Adrenaline infusion acts to rescue the adrenalectomized dog suffering from bleeding, i.e. the animal survives a severe hemorrhage. This corresponds well with the statement re the effect of medulli-adrenalectomy upon the life and the blood pressure fall after hemorrhage. As to the view that in considering the question of functions of the medulla, it should be kept in mind that no important functions can in any case be assigned to an organ the loss of which causes no symptoms,[7] we cannot concur.

In rabbits the double splanchnicotomy also considerably reduces the blood sugar increase due to hemorrhage, whereas double adrenectomy reduces it to a lesser degree.[8,9] Splanchnicotomy can almost prevent a reduction of adrenaline content of rabbit adrenals by bleeding.[10]

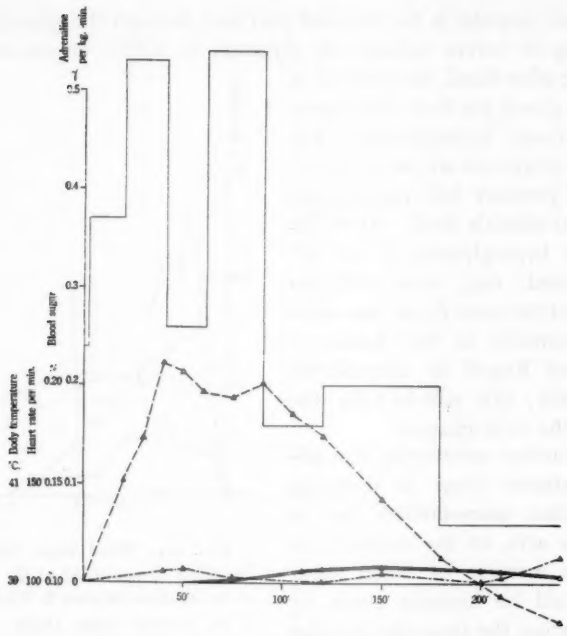

FIG. 13. Blood sugar fluctuation in doubly splanchnicotomized dogs, in imitating the oversecretion of adrenaline causable by bleeding one-third of the total blood volume. (Tachi & Saito)

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7) Stewart, *Arch. of Int. Med.*, 1929, **43**, p. 765.
8) Tachi, *Tohoku J. Exp. Med.*, 1928, **10**, 96.
9) *Id.*, *Ibid.*, 307.
10) *Id.*, *Ibid.*, 409.

PEPTONE

Peptone, in an intravenous dose of 0.1–0.3 g. per kg., is capable of accelerating the adrenaline secretion in the normal dog; Type B_1. On an average, the maximum rate is 0.4 γ per kg.-min., which appears ten minutes after injection, the oversecretion lasts for thirty minutes, and the total amount

of oversecretion 12 γ per kg., schematically stated.

If the spinal cord is cut before the thoracic cord, 0.05–0.1 g. per kg. peptone acts also to accelerate the adrenaline secretion, but this time it is small; three to ten times acceleration is commonly observable, but 0.1 γ per kg.-min. was also observable. A greater dose of the drug can not be tried because of the weakness of dogs so operated on.[2]

What part is played in this oversecretion in peptone intoxication by the central mechanism located cranially is difficult to elucidate from the data given.

Ether narcosis depresses the action of peptone upon the adrenaline secretion, and finally it can nullify the action.[1]

It is noteworthy that the oversecretion of adrenaline, if any, takes place so long as the depressed state of the animal is perceivable.[1]

In a dog (No. 34) among 7, in which 0.3 g. per kg. was dosaged, a considerable oversecretion of adrenaline was estimated: the maximum rate 2.4 γ per kg.-min. at 10 minutes after injection, the period of oversecretion over five hours, and the whole amount 200 γ per kg.[1] Adrenalinemia of a similar scale was also detectable in bleeding experiments, though infrequently.

The oversecretion of adrenaline by peptone results through the splanchnic nerves.[3]

Giving 0.2–0.3 g. peptone per kg. produces a hyperglycemia of such a scale: the maximum 0.15–0.2% in 20–30 minutes after injection, the hyperglycemic period 1.5 hours and the excess sugar 3 (1.5–4.5) g. Five minutes after injection the sugar content starts to increase. In dog 34 of Watanabe, given above, hyperglycemia was also large and of a long duration (0.2%, 10 g.).

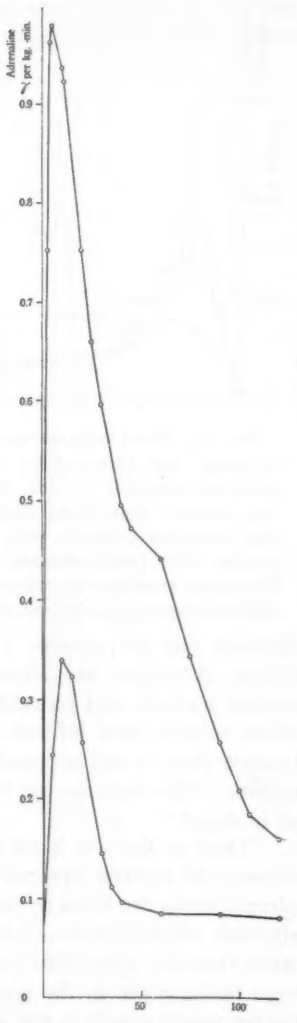

FIG. 14. Adrenaline secretion on peptone (0.1–0.3 g. per kg.) (Lower curve, Watanabe) and on anaphylactic shock (Upper curve, Sato, Ohguri & Wada).

Peptone hyperglycemia in dogs is reduced by splanchnicotomy as well as by medulli-adrenalectomy; schematically the reduction is 20% by the former, and 40% by the latter.[3,4] The

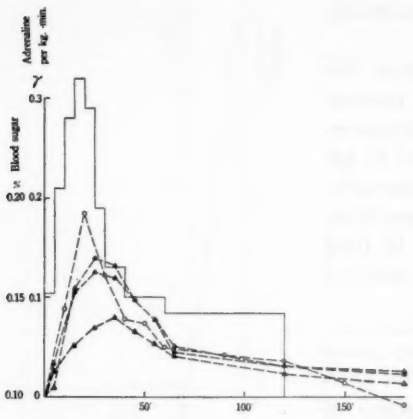

FIG. 15. Blood sugar fluctuation on peptone in normal dogs (light circles) and in doubly splanchnicotomized. In the latter group of dogs, peptone alone (light triangles), peptone plus adrenaline (triangles with point) or adrenaline alone (solid triangles) was given i.v. Histograms show how adrenaline hydrochloride saline solution was i.v. infused. (Kaiwa)

upper sympathetic ganglia in the abdominal cavity was removed in addition to the medulli-adrenalectomy, with about the same outcome.[5]

Adrenaline-saline solution, intravenouly introduced in the splanchnicotomized dog in imitating the secretion in the peptone poisoning in the normal dog is capable of causing hyperglycemia of a certain degree, but it is definitely inferior to one causable by peptone in the splanchnicotomized dogs. And if both the injections, peptone and adrenaline, be combined, the glycemic effect in the dog so operated on is almost similar to that of peptone alone.[4]

Similar tendencies are also obtainable in rabbits re the glycemia due to peptone, i.e. influence of splanchnicotomy and adrenalectomy thereupon are almost identical with those in dogs. In dogs the adrenal medulla and in rabbits the adrenals were removed. Adrenaline-saline solution was infused intravenously here in a dose taken at random, because there is still no method to estimate the adrenaline secretion rate in rabbits. The outcome can be taken as not much different from that obtained in dogs.[5,6]

Thus on the one hand ether anesthesia diminishes or abolishes the occurrence of peptone hyperadrenalinemia, while on the other hand the hyperadrenalinemia produced by peptone does not act, at least, to further the hyperglycemia which is produced in the peptone intoxication through the mechanism other than the adrenaline oversecretion. If the oversecretion of adrenaline were taking place in the peptone intoxication in the dog under ether, what figures would result in the matters under test, i.e. those commonly altered by the action of adrenaline? Experiments done with such thoughts in mind show that the adrenaline infusion produces only its usual effects on the arterial pressure and pulse rate, but no definite relationship was established re the blood sugar concentration and the body temperature.[7]

Here it may be mentioned that in dogs the glycemic effect of histamine is not altered by adjusting the rapidity with which the drug is applied in-

travenously,[8] contrary to the case with peptone and histamine on rabbits.[9]

Peptone, intravenously applied, slightly reduces the adrenaline content in rabbits, irrespective of whether the splanchnic nerves are cut or not. When death occurs there very rapidly, no reduction takes place.[10]

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HISTAMINE

Histamine produces an oversecretion of adrenaline in a form similar to peptone, namely of Type B_1, and its degree depends upon doses. The impulse is sent through the splanchnic nerves to the adrenal medulla.[1]

It was found that 1 mg., 2 mg. and 4 mg. histamine per kg. causes an oversecretion with the maximum rate of 0.4, 0.6 & 1 γ per kg.-min. respectively. These appeared 5 minutes after injection; the rate decreased rapidly at first, and the oversecretion almost disappeared 40 minutes to 2 hours later. The total amount of oversecretion was 10, 30, & 45 γ, mean 30 γ per kg. of body weight.

Blood pressure fell rapidly, the maximum fall by 60–80 per cent reached 5 minutes after injection. The pressure fall and the adrenaline secretion increase go quite simultaneously. It is impossible to determine which precedes the other, so far as the methods adapted show. The blood pressure curve recovered somewhat slowly, but the time occupied for the complete

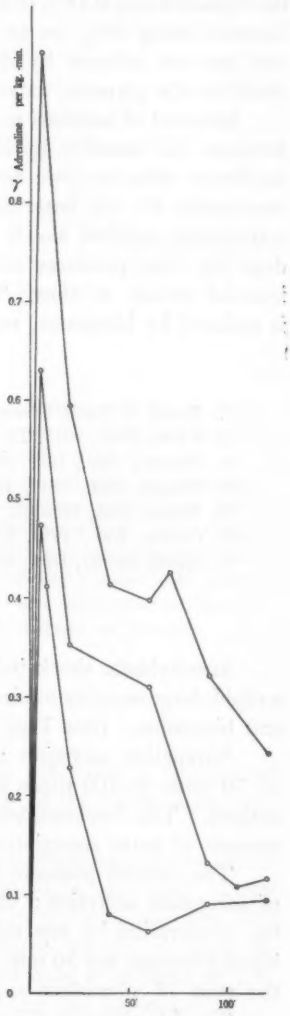

FIG. 16. Adrenaline secretion on i.v. histamine. (Wada et al.) Upper curve—5 to 3 mg. histamine per kg.; middle—2 mg. & lower—1 mg.

recovery was almost identical for both events.

The blood sugar content reached the acme 20 minutes after injection, the highest being 0.15% (0.13–0.2%). The heart beats rapidly, the maximum increase being 50% on an average. Changes in blood pressure and pulse rate are not affected by double splanchnicotomy, while neurotomy nearly abolishes the glycemic action of histamine.

Removal of adrenals in rabbits has also no material effect upon the blood pressure fall causable by histamine.[2] It acts to only slightly decrease the minimum effective dose and the lethal dose as well.[3] The medulla is not responsible for the latter fact.[4] Further it may be added: histamine, intravenously injected slowly to rabbits, always produces hypoglycemia.[5] In dogs the drug produces only hyperglycemia, irrespective of whether it is injected rapidly or slowly.[6] The adrenaline content of adrenals in rabbits is reduced by histamine, regardless of the speed of injection.[7]

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Anaphylactic Shock

Anaphylactic shock induces in the dog, non-fastened, non-anesthetized, a rapid, large acceleration in the adrenaline secretion of Type B_2, as do peptone and histamine. (See Figs 14 & 16)

Adrenaline secretion reaches its peak, mean 1 γ per kg.-min., about 20–30 times to 100 times the initial rate, 5–10 minutes after the injection of antigen. The hypersecretion lasts for two hours or longer, and the total amount of extra secretion 45 γ per kg. on an average.[1]

The arterial pressure falls almost simultaneously with the acceleration of adrenaline secretion; there was a case witnessed where the fall preceded the acceleration by one minute, among six cases. The lowest level in the blood pressure, say 30 mm. Hg., came simultaneously or some minutes before the peak of adrenaline secretion.[1,2]

The blood sugar did not begin to elevate until a few minutes after the above two events, and its peak appeared commonly 10 to 30 minutes after injection. The hyperglycemic period is sometimes preceded by a transitory hypoglycemia of a very small scale; the hyperglycemic phase covers 100 minutes on an average.

Double splanchnicotomy as well as double medulli-adrenalectomy reduce the intensity of anaphylaxia hyperglycemia; the former acts weakly and the latter strongly. Roughly speaking, the former reduces it by twenty per cent, the latter by forty.[3] Both operations have no material influence upon the severity of blood pressure fall, but the recovery course seems somewhat slow.[3]

When the hyperglycemia starts, the dog is already in a depressant state, and the blood pressure begins to recover from the fall.

When the poisoning is too severe, profound depression develops accompanied by low pressure and low sugar, and eventually death takes place.

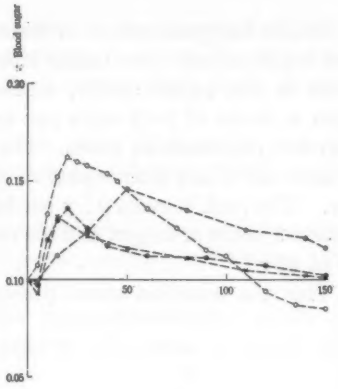

FIG. 17. Blood sugar fluctuation on anaphylactic shock. (Ohguri; Ohguri; Sato, Ohguri & Wada; Ohguri)

Shock in normal dogs (light circles), and in medulli-adrenalectomized (solid circles). Adrenaline hydrochloride saline solution infusion in medulli-adrenalectomized animals (light circles with point), and shock and adrenaline infusion in the dogs so operated on (crosses).

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CAROTID SINUS NERVE

Clamping of the common carotid arteries does not effect in the normal dog any increase in the adrenaline secretion rate or of the blood sugar concentration.[1]

In the cava pocket experiment in the dog under ether, stimulation of the H.E. Hering nerve causes a reduction in the adrenaline secretion. Forty seconds after the start of the stimulation a reduction becomes manifest, and 10–30 minutes are required after stopping stimulation for the complete recovery of the rate. On a 2 minute stimulation the rate is reduced to 1/4-1/3 of the initial, or from 0.5 γ per kg.-min. to 0.15 γ per kg.-min.[2]

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Insulin

Insulin brings about an oversecretion of adrenaline of B_1 Type. If the blood sugar content goes under 0.06%, the oversecretion of adrenaline sets in, but at first rather slowly, so that 20 minutes after the i.v. injection of insulin in doses of 3–10 units per kg. an oversecretion is now unmistakably observable in almost all cases. The oversecretion reaches its peak one and one hour and a half after injection, and then diminishes slowly to the initial value. The peak is mean 0.5 γ per kg.-min. (0.2–1.5 γ), the duration of oversecretion 3 hours or longer, and the total amount 65 γ per kg., with the greatest case of 80 γ.[1]

That the secretion curve presented here has a plateau should not be

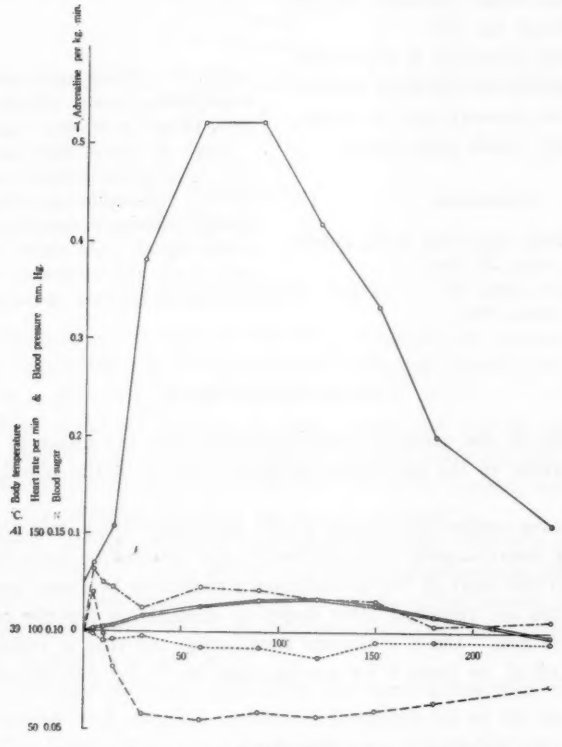

Fig. 18. Insulin, i.v. 3–5–10 units per kg. Adrenaline secretion, blood sugar, blood pressure, pulse rate & anal temperature. (Yen, Aomura & Inaba)

taken as a characteristic of insulin, because each curve has a sharp peak commonly.

This action of insulin is also producible in the depancreatomized dog. It should be remembered that the oversecretion is also produced before the blood sugar concentration reaches the limit above noted, if the decrease is taking place very rapidly.[2]

By way of precaution it may be here added that insulin acts in the pancreatomized animal to cause the blood sugar fall more readily and more rapidly in comparison with the control.

Insulin loses almost or entirely its hyperadrenalinemic action on the dog, after the spinal cord is cut before the thoracic, contrary to bleeding, peptone or sensory stimulation.[3]

Splanchnicotomy wholly annuls the hyperadrenalinemic ability of insulin. Neurotomy increases the susceptibility of animals to insulin and its hypoglycemic power.[4]

Hypnotics, such as avertin,[5] evipan,[6] do not materially interfere with the occurrence and magnitude of insulin hyperadrenalinemia. The blood sugar decreases there considerably, the general blood pressure too, but clinical symptoms such as tremors and convulsions seldom occur, as is to be expected.

The data in rabbits,[7] that the hypoglycemic action of insulin is retarded distinctly by a large dose of barbital apparently disharmonize with the above mentioned. Is it to be explained by the different kinds of animals experimented on, of the drugs, or of the amount of drugs applied? A smaller dose of barbital does not cause any retardation.

Insulin reduces the adrenaline content of adrenals in rabbits. Splanchnicotomy annuls this effect. Intravenous introduction of glycose also acts likewise.[8] A repeated insulin application to rabbits for a long period, i.g. five weeks, causes an increase of the adrenaline load. It depends upon the intact innervation. If insulin is given i.v., the adrenaline load in adrenals increases slightly and quite transitorily.[9]

The abolition of adrenalin secretion, i.e. by demedullation of the adrenals, exaggerates in dogs the intensity of insulin hyperglycemia. And the intravenous infusion of adrenaline-saline solution, conducted in a similar manner and magnitude as normally occurring in the insulin poisoning, acts to compensate the medulli-adrenalectomy in respect to the blood sugar fluctuation, if done in addition to the injection into the dog deprived of the adrenal medulla.[10]

There are some divergent reports re the relation between the adrenalectomy[11] or splanchnicotomy and the glycemic action of insulin in rabbits. Either no alteration,[12] an earlier appearance of hypoglycemia[13] or a largeness of the latter.[11]

Insulin acts to decrease the body temperature definitely though slightly when applied to the medulli-adrenalectomized dog; and it is well compensated

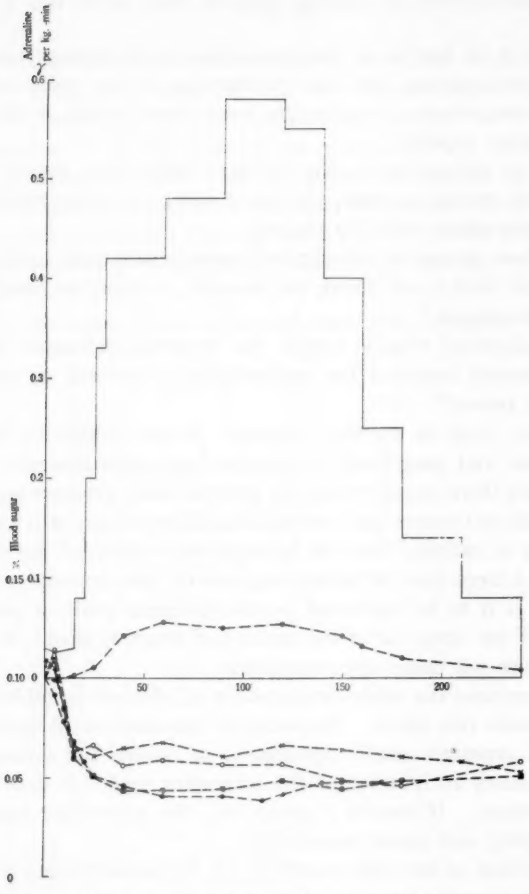

FIG. 19. Blood sugar fluctuation on insulin, 5 units per kg. i.v. (Saizyo)

Insulin in normal dogs (light circles), and medulli-adrenalectomized (solid circles). Infusion of adrenaline solution, as shown in histograms, in medulli-adrenalectomized dogs (light circles with point) and both chemicals combined in those so operated on (crosses). Infusion of saline solution combined with insulin injection (double quadrates).

by the introduction of adrenaline solution in amounts and course corresponding to the adrenaline secretion caused by insulin in the normal animal.[10]

Contrary to the results given above, the infusion of adrenaline solution carried out in similar experimental conditions has little or no effect on the

arterial pressure or the pulse rate; the reverse action was never discovered on the other hand.[10]

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GUANIDINE

Guanidine is a potent drug in accelerating the adrenaline secretion, like insulin.[1] Its course is of B_2. However there are some different figures; first, the guanidine hyperadrenalinemia develops in the normal dog soon after the injection, and more rapidly than in the case with insulin. Twenty minutes after the injection of guanidine the hyperadrenalinemia reaches the peak 0.6 γ per kg. per minute, 10 times acceleration on an average on intravenous injection of 0.15 to 0.3 g. per kg. guanidine hydrochloride. Secondly, guanidine causes first a small but long hyperglycemic period, which is replaced by the development of hypoglycemia, arrived at 0.04% 100 minutes further later, while the adrenaline acceleration is of quite the same scale in both sets of experiments.

The oversecretion period covers three hours or longer, and the total amount secreted is computed as 50 γ per kg. (in 160 minutes), figures similar to the insulin experiments, given in doses of 3 to 10 units.

When excessive dyspnoea develops a copious oversecretion breaks out. The thick lines in Fig. 20 indicates the two cases with dyspnoea or opisthotonos, whereas the other two cases without dyspnoea are indicated by the thin lines.

Splanchnicotomy deprives the hyperglycemic ability from guanidine. Hypoglycemia of a similar depth as in the normal individual takes place there however.[2]

The time relation of the adrenalinemia and the glycemia is thus different depending upon the insulin and the guanidine. In the case of insulin the oversecretion of adrenaline can be accounted for the rapid and large development of hypoglycemia, but in guanidine poisoning the adrenaline over-

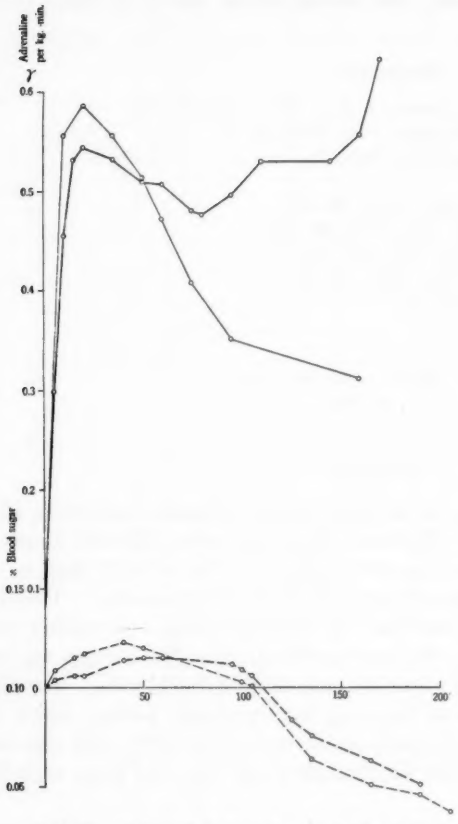

FIG. 20. Adrenaline secretion and blood sugar on guanidine, 0.15 to 0.3 g. per kg. i.v. (Sugawara & Tada)

Thick lines originate from 4 cases in all; the two cases without dyspnoea are indicated with thin lines.

secretion is already undergoing some reduction at the time when hypoglycemia first makes its appearance. The hyperadrenalinemic action of guanidine must be due to a mechanism other than that for insulin.

In the doubly splanchnicotomized dog, guanidine was injected i.v. and adrenaline hydrochloride-saline solution was also injected i.v. at the rate of secretion, usually presents after guanidine. This procedure produces a hyperglycemia on a scale, standing just between those brought about by each injection when given singly. The hyperglycemia obtainable in the normal dog takes a mediate course between that causable in the splanchnicotomized dog by guanidine alone and that by guanidine and adrenaline. The pattern which was applied here was just that with the greatest acceleration.

In rabbits guanidine (0.24–4 g. per kg. subcutaneously applied) produces a hyperglycemia lasting for some hours in about two thirds of the cases, while a hypoglycemia takes place in all cases, with or without a foregoing hyperglycemia. Splanchnicotomy reduces the hyperglycemia, while adrenalectomy can nullify it entirely. The latter operation reduces the hyperglycemic ability of synthaline there.

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Sensory Stimulation

(Fastening, Emotion and Sleep)

Sensory stimulation evokes generally an oversecretion of adrenaline. Usually the acceleration observable in unanesthetized dogs is definitely small compared with other known agencies. In our experiments the strength of faradic stimulation was taken usually as to cause violent aggressive reaction in the animals.

The height of oversecretion due to the stimulation depends upon the preliminary pre-existing rate. Generally speaking, the reflex stimulation increases the secretion by two and a half to three times commonly, irrespective of varied pre-existing rates. The following table illustrates this:

Table XI

Influence of Sensory Stimulation upon Adrenaline Secretion Rate in Dogs

(per kg. of body weight and per minute)

	Pre-existing rate	Increase	Ratio
No-narcosis, lumbar route method[1]	0.07 γ	0.1 γ	1 : 2.5
No-narcosis, cava pocket method[2,3]	0.65 γ	1 γ	1 : 2.5
Ether narcosis, cava pocket method			
Kodama[4]	0.8 γ	1 γ	1 : 2.3
Sugawara et al.[3]	0.5 γ	0.4 γ	1 : 1.8
Stewart & Rogoff[5]	0.2 γ	0 γ	1 : 1

Narcosis masks the augmenting effect of sensory stimulation on adrenaline secretion; the fact that accomplished physiologists such as Drs Stewart and Rogoff failed to find any effect at all is due to a too deep anesthesia indeed.

The smallness of the pre-exsting rate in the absolute quantity as well as in the limits of fluctuations in the reports from the Cushing Laboratory in Cleveland indicate the deepness of anesthesia. They regard the former as the condition favourable for an increase of secretion and the latter as a proof of the correctness of their cava pocket method. Just the reverse is true, however. They used, it may be added, at that time ether, urethane, etc.

Stimulation was conducted in our cases for 20 or 30 seconds, and we found that the acceleration took place not only during the excitation, but after the cessation in certain instances, as was proved by some samples collected from 5 seconds after the cessation of stimulation—so long the after-action of sensory stimulation.

The greatest acceleration of adrenalin secretion in normal dogs (eight in number),[1] caused by artificial electrical stimulation of a nerve branch was 2 to 5 times, seldom 8 times the pre-existing value, or 0.02 γ–0.2 γ from one

gland, i.e. 0.2 γ per kg.-min. from both glands as the mean. We had the following remarkable experiment: on fastening, the dog (No. 7) cried and struggled very furiously and uninterruptedly throughout fastening. The left lumbar route operation was carried out meanwhile with great difficulty; the adrenal vein blood samples collected twenty minutes later revealed an enormous rate of secretion, such as 6 γ per kg.-min., i.e. 100 times the basal rate. Such a great acceleration has been obtained in dogs only with nicotine (200 times), CO and KCl.

We are still not in a position to elucidate, how the emotional factor is involved in this kind of experiment, like fastening, sensory stimulation, etc. We were at that time very strongly inclined to believe an exceedingly great role is played by the emotional factor in the extraordinary oversecretion of adrenaline just illustrated.

Fastening a normal dog on an operation table, for example, induces a hyperadrenalinemia, though slight and not constantly. About two times acceleration occurs in half of the cases.[1]

Fastening glycemia is a classical fact. While it always occurs in cats and rabbits, it is not constantly producible in dogs. The latter fact is in harmony with the adrenaline matter in that kind of animal. How the emotional element intervenes in producing the fastening diabetes has been a much discussed subject for a long time.*

Sensory hyperadrenalinemia occurs also in the dog, the spinal cord of which is sectioned between the cervical region and the thoracic. But the effect is definitely weak, and only seldom does the acceleration take place, then rather small, i.e. at most three times the initial rate. A great contrast to bleeding or peptone.[6]

Tests show that sensory stimulation, which always produces a paradoxical pupil reaction in normal cats (0.5–2 mm. dilatation), has a weak effect after double splanchnicotomy (in 8 out of ten; 1 mm. in three), and weaker in doubly medulli-adrenalectomized (in 6 out of 10; 0.5 mm. in six). The barking dog is very weak in producing a similar effect; after each operation the dilatation occurs no more.[7]

Hyperglycemic effect in rabbits subjected to sensory stimulation is largely reduced by double splanchnicotomy, while double adrenalectomy is incapable of doing this.[8] The latter fact almost harmonizes with the fact that sensory stimulation contracts in an anesthetized cat a limb or spleen denervated, regardless of whether the adrenals are removed or not.[9]

During sleep the adrenaline secretion diminishes from the basal rest rate, for example, from 0.02 γ per kg.-min. from one adrenal to 0.01 γ.[1]

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* Cf. I. Fujii, *Tohoku J. Exp. Med.*, 1920, 2, 9. When Fujii's paper was published, Dr. W. B. Cannon wrote to me personally saying that the data that the intensity of fastening hyperglycemia varies daily, and in fact does so a form of wave, indicates its emotional origin.

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PIQÛRE DIABETIQUE

On puncturing the floor of the fourth ventricle, according to Bernard, the adrenaline secretion increase rapidly;[1] in five minutes the rate reaches commonly a high value such as 0.5 γ per kg.-min., and further increases though slowly from then on. The peak, such as 0.2 γ to 1 γ is noted thirty minutes after puncturing, then the rate diminishes slowly; B_1-type. The total over-secretion is mean 50 γ per kg., and the greatest 90 γ; 13 γ in the first half hour, 12 γ in the second half hour, 15 γ in the second hour, and 12 γ in the third. Roughly stated, the piqûre increases the adrenaline secretion fifteen times in the rate and five times in the total amount, the whole period of augmentation being over three hours.

The blood flow through the adrenals increases here moderately or slightly; it never decreases. The increase of secretion depends, however, chiefly upon that of the concentration of hormone.

On a closer examination, the blood sugar increase precedes the adrenaline secretion acceleration in some cases, sometimes occurs simultaneously, and in still other cases it follows the acceleration. Usually the peak appears earlier by some minutes in the glycemic curve. Hyperglycemia is noted as 0.15 to 0.2%, mean 0.16%.

Ether anesthesia depresses the increase in both events, but the blood sugar content suffers largely therefrom, so that the hyperglycemia is missed frequently. On the contrary, the adrenaline secretion commonly increases, though on a smaller scale than in non-anesthetized dogs. From 1.5 to 3 times acceleration is common. The pre-existing rate of secretion is 0.1 γ per kg.-min. for one gland in a dog under ether, equipped with the lumbar route preparation. Blood pressure fluctuation, observable after the piqûre, also, suffers from narcosis, as explained below.

Piqûre produces hyperglycemia in the dog, deprived of either adrenals or medulli-adrenals, but on a somewhat smaller scale than in the normal individuals. The former operation was carried out acutely in the de-afferented dog; accordingly, neither fastening nor narcosis was needed.[2,3] The

FIG. 21

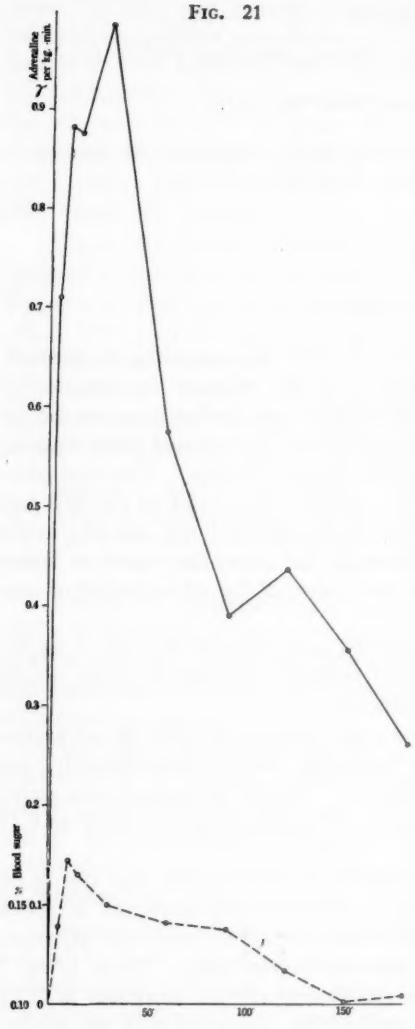

FIG. 21. Adrenaline secretion and blood sugar content on piqûre diabétique. (Yen, Kaiwa & Wada)

FIG. 22

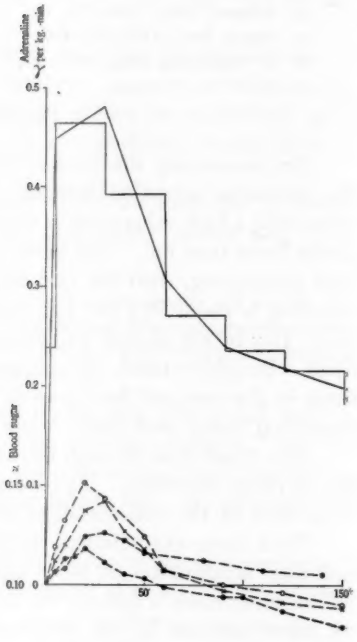

FIG. 22. Blood sugar fluctuation on piqûre diabétique. (Yen, Kaiwa & Wada; Wada & Kaiwa)

Piqûre in normal dogs (light circles), in medulli-adrenalectomized or adrenalectomized dogs (solid circles), adrenaline hydrochloride-saline solution infusion to such dogs (light circles with point), and both agents combined in such dogs (crosses).

excess is of one-half of the normal cases; 0.12% against 0.15%. And this inferiority is nearly compensated by infusing adrenaline-saline solution intravenously in the scale similar to the natural secretion of adrenaline in the

normal dog, punctured according to Bernard. Fig. 22 illustrates this.

In these respects the rabbit affords some different figures from those above. Double splanchnicotomy,[4] double adrenalectomy or double medulli-adrenalectomy reduces the magnitude of piqûre hyperglycemia largely. While the piqûre hyperglycemia is of such a scale as 0.25% in normal individuals, three kinds of operation reduce it to 0.12%.[5] The figures of the Cleveland physiologists differ widely from those above referred to; the glycemic height before and after adrenalectomy is almost equal, as 0.41% against 0.45%.[6]

It will be in order to mention here the piqûre experiments with the devervated heart in cats[7] as well as with the denervated pupil in rabbits,[8] in which double adrenalectomy abolishes the paradoxical reaction.

The arterial pressure either ascends or descends abruptly on a large scale on puncturing the fourth ventricle in the normal dog, but soon starts to descend or ascend as the case may be, reaching the initial level about ten minutes after puncture, roughly speaking. Afterwards the pressure fluctuates commonly under the initial level on a small scale. Double medulli-adrenalectomy, with or without accompanying double vagotomy, has some tendency to exaggerate the fall in the later period, and the adrenaline infusion acts to compensate it, though never significantly. Blood pressure fluctuations on the piqûre suffer from narcosis in a similar manner to the adrenaline secretion.

The pulse rate diminishes on puncturing suddenly and largely, approximately 50%, and then at first rapidly and later slowly it tends to recover. In the piqûre experiments it is difficult to elucidate the role of adrenaline oversecretion in the pulse frequency variations. In a few cases the pulse rate increases rapidly, but soon diminishes. No peculiar characteristics show themselves in the further course.[1,2,3]

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Cerebellar Puncture

An injury of the cerebellum such as destroy the whole lobulus posterior medianus, particularly the vermis, results in an oversecretion of adrenaline and an increase of blood sugar content. The former is similar in form to

that due to piqûre diabétique, but on a decidedly small scale, say one-third in the maximum rate as well as in the total amount; they are 0.15 to 0.35 γ per kg.-min. and 12 γ per kg. respectively.

The blood sugar increase is of much small scale; the greatest excess 0.02%. Blood pressure soon ascends largely on puncturing, but soon to descend and in 10 to 20 minutes completely recovers the initial level.

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MUSCULAR EXERCISE (EXHAUSTION)

Muscular exercise, as running, for example, on a paved road or in a revolving wheel, does not augument the adrenaline secretion at all in the case of the dog not yet exhausted. When much tired or exhausted, an increase sets in, but only on a small scale, the peak being noted a few minutes to a half-hour after ceasing to work. The maximum rate is mean 0.08 γ per kg.-min., the greatest case being 0.2 γ. The oversecretion lasts for about one hour.[1]

The blood sugar content fluctuates only insignificantly; it is apt afterwards to decrease though slightly. Extirpation of the adrenal medullae, cutting of the splanchnic nerves and their combination do not interfere with the scale of decrease.[2]

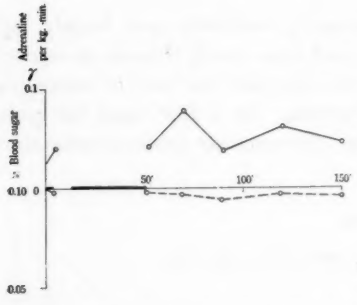

FIG. 23. Adrenaline secretion and blood sugar content on muscular exercise. (Wada, Seo & Abe)

Thick lines on the abscissa show the duration of running. First running: 380 m. in 3 min. No sign of fatigue, pupils dilated slightly. Second running: 684 m. in 39 min. Refused several times to continue to run. Panting occurred at times. At the end of running seemed to be tired, but not exhausted.

Muscular exhaustion does not effect to reduce the adrenaline content of adrenals in rabbits, irrespective of whether the innervation be impaired or not.[3]

Normal and doubly medulli-adrenalectomized rabbits behave quite the same re the fatigability of the nerve-muscular preparation *in situ*, whereas double adrenalectomy shortens it to one-third, 10 hours against 30 hours.[4]

While these three kinds of experiments well harmonize with the above given data re the adrenaline secretion on muscular exercise in the normal dog, there are however other sets of researches which completely disharmonize with them.

They are those of Hartman and

of Cannon with the denervated pupil or heart in the cat. The paradoxical reaction occurs on muscular work. When the adrenal medullae be put out of function the reaction does no more occur.

The paradoxical pupil reaction in cats, conducted in our laboratory partly confirmed the outcome of Hartman. It was observable in a treadmill experiment.[5] A travel of over 200 meters caused fatigue. The pupil dilated by 2 mm. commonly, which disappeared a few minutes later. The pupil started to dilate before any sign of fatigue had set in. Removal of the adrenal medullae or double section of splanchnic nerves much reduces the paradoxical reaction in such a scale as 0.5 mm. dilatation in both cases against 2.5 mm. in the control. In some cases no reaction was observed.

That the paradoxical pupil reaction occurs on a definitely smaller scale after the medulli-adrenal function is ruled out indicates undoubtedly that the reaction that takes place during the muscular work depends largely upon the intergrity of the medulli-adrenal function.

The findings that no adrenaline oversecretion is determinable during muscular excercise by the direct method in the normal animal, while the paradoxical pupil reaction which in a large extent depends on the medulli-adrenal overaction takes place meanwhile, remain to be analyzed carefully. The query will be dealt with further in the next chapter.

By way of precaution: in the adrenaline secretion experiment no adrenal venous blood sample was obtained during the muscular work. To collect it, the latter must be discontinued. The samples taken immediately after the discontinuance showed no oversecretion, but later an oversecreion was discovered to start and progress further, when the animal was exhausted, as Fig. 25 shows. The fact led us to assume that there was no oversecretion of adrenaline during the muscular exercise, as above.

The data re leucocytes number and pulse rate in the totally sympathectomized dog[6] well harmonize with the paradoxical pupil reaction in cats of Itikawa especially. Vigorous excitement and walking brings an increase in leucocytes number by about 100%, and the total sympathectomy brings the excess down to 20%. Pulse rate is accelerated by 20–40% on vigorous excitement and walking, and the acceleration is also largely reduced by total sympathectomy. Provided the body temperature elevation be taken into account the acceleration is computed at 10–20%. The red blood cell count remains unaltered by such manipulations throughout.

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4) Sibuta, *Ibid.*, 1944, **46**, 334; Oouti, *Ibid.*, 1945, **48**, 259.
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6) Fuzii, *Ibid.*, 1938, **34**, 542; The data are in accord with Brouha, Cannon and Dill, *J. Physiol.*, 1936, **87**, 345.

Cooling

Application of cold to the animal organism can produce an elevation of adrenaline secretion rate, provided the severness of cooling reaches a certain limit. It is applied directly to the body surface or by introducing cold fluid into the stomach; in the latter case it is safe to carry it through the stomach fistula, previously prepared, so as to avoid emotional disturbances that might vitiate the experimental outcome.

On dipping the normal dog into cold water of 0.5°–1°C. for twenty to forty minutes, so that the anal temperature falls to thirty degrees or thereabouts, the adrenaline secretion increases always. The rate, measured immediately after withdrawal from the bath, is the peak, mean 0.3 γ per kg.-min., and then the rate rather rapidly diminishes. But the whole course might be taken as Type B_1. The oversecretion extends for about 100 minutes and its total quantity is 13 γ per kg.[1]

In these experiments the duration of dipping was so taken as to cause a similar degree of anal temperature fall; the maximum rate in the adrenaline secretion, its total amount, and pulse rate variation were almost uniform throughout most cases, while the blood sugar increase varied individually.

Fig. 24 shows the general course of these factors.

Body temperature fell to 28°C. (30.5° to 25°) After the withdrawal the temperature descended further for some time; the above figures show the lowest.

When the body temperature falls by 0.5°–1.7°C. by subjecting the dog to superficial cold, as when putting it into a rubber bag, containing cold water at 5° to 7°C., or placing the animal on a block of ice for half an hour, no acceleration is detectable in the adrenaline secretion at all.[2]

Introduction of ice-water at 5°C. in an amount of 1 000 to 1 500 cc., as to occasion a heat-debt of 2 000 to 3 500 calories, causes a small cold stimulus, probably the smallest, to bring about an oversecretion of adrenaline. The highest rate arrived at 0.1 γ per kg.-min. (0.05 γ–0.2 γ) and the lowest anal temperature mean 38°, 37.5° being the actual lowest.[2]

The cold hyperglycemia occurs, generally speaking, in parallel with the oversecretion of adrénaline. And the upper limit of low temperature for eliciting both disturbances is practically the same; moreover, the course of hyperglycemia and of hyperadrenalinemia coincide well with each other. Figs. 24 & 25 illustrate this.

In most cases peaks of both the events were estimated at the same time; in one case only the peak appeared earlier in the adrenaline secretion, and the reverse in another case. Unless the collection of blood samples be done with much smaller intervals, no such definiteness can be said of the exact time relation however.

When in consequence of the dog being dipped into the cold bath the body temperature fell under 30°C., the peak of hyperglycemia appeared 10 to 20 minutes after withdrawal therefrom, and was estimated as mean 0.17% (0.16 to 0.2% on an average) and hyperglycemic period extended for 100 to 200 minutes on an average. The extra sugar for the period is thus 3 mg. per kg. on an average.[1,3,4]

When cold water was poured into the stomach through the gastric cannula, so as to result in a small fall of body temperature as by 1°C., the scale of the hyperglycemia was small, as 0.115% and 0.56 g. extra sugar.[1]

FIG. 24

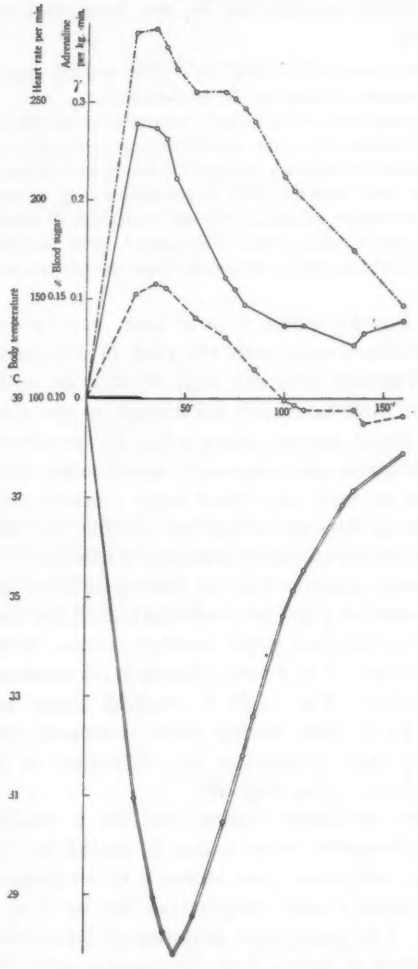

FIG. 24. Adrenaline secretion (solid line), blood sugar content (broken line), heart rate (chain line) and anal temperature (double line) on cold application to dogs by means of cold bath (about 1°C). (Wada, Seo & Abe)

The time in the cold bath is shown with thick line on the abscissa.

FIG. 25

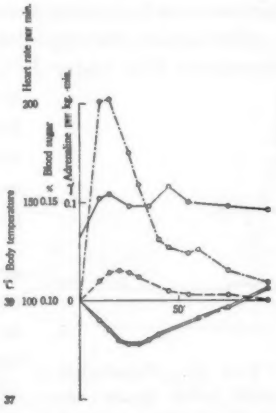

FIG. 25. Introduction of cold water of 5°C. into the stomach of the dog through the gastric fistula. (Wada, Seo & Abe)

The legend as in Fig. 24.

Demedullation of adrenals usually reduces the magnitude of cold-diabetes, though insignificantly; Cf. Figs. 7 & 8. Occasionally no reduction was estimable. It may be rather superfluous to say here that the cold-diabetes is one of central origin.

Increase of metabolism[5,6] as well as occurrence of shivering[7] on cooling are not significantly altered by de-medullation of the adrenals, similar to the cold-diabetes.

It might be noticed in passing that introduction of cold water, for example, should not be made through the esophagus, else the passage of a tube itself produces hyperglycemia of a certain strength. Even if it is poured into the stomach through the fistula as to produce a fall of 1° to 2°C. in anal temperature, a small hyperglycemia is producible only in male animals in which emotional expression is somewhat distinct, while an occurrence of hyperglycemia is ambiguous in female animals which behave there quite calm.[8] The fact that occurrence of emotional reaction as well as diabetes differs depending upon the sex has long been well known.[9]

Body temperature reaches its bottom either a little later, say by ten minutes or thereabouts, than or simultaneously with the peak of adrenaline secretion and of hyperglycemia. The fall does not stop at once on withdrawal from the cold bath. Two or three hours are not enough to see a recovery of anal temperature to the initial degree, when a big fall as low as 30°C. is brought about. Its complete recovery commonly would come some ten minutes later than the adrenaline secretion and blood sugar concentration.

The paradoxical pupil reaction is brought about on cooling the dog body in a cold bath, and it is minimalized by exclusion of adrenal medullae.[8,10,11] This outcome harmonized well or nearly entirely with the finding of Hartman.

On dipping the dog with a denervated pupil in a cold bath until the anal temperature falls under 30°C., the paradoxical pupil reaction occurs, on an average 6 mm. (4 to 8 mm.) dilatation *ad maximum* being noted. The acme is reached about ten minutes or so after having been immersed, and one hour after immersion the dilatation is no more visible.[3] (See Fig. 26)

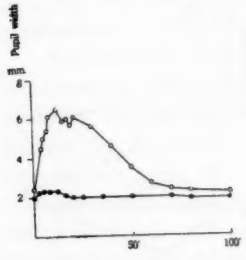

FIG. 26. Paradoxical pupil width (g.c.s. excised) of dogs in cold bath experiments. (Itikawa)

Light circles—with intact adrenals; solid circles—after medulli-adrenalectomy.

Quite the same finding, but on a smaller scale is obtainable when a dog is cooled by introducing cold water into stomach in an amount as to occasion a body temperature fall by 2 to 3 degrees. The paradoxical dilatation of 2 to 3 mm. *ad maximum* is noted 5 to 10 minutes after the introduction, and thirty minutes after the introduction the pupil recovers its initial width.[10] (See Fig. 27)

These paradoxical pupil dilatations are almost annulled by the demedullation of adrenals, but a trace is still detectable however. It is thus not

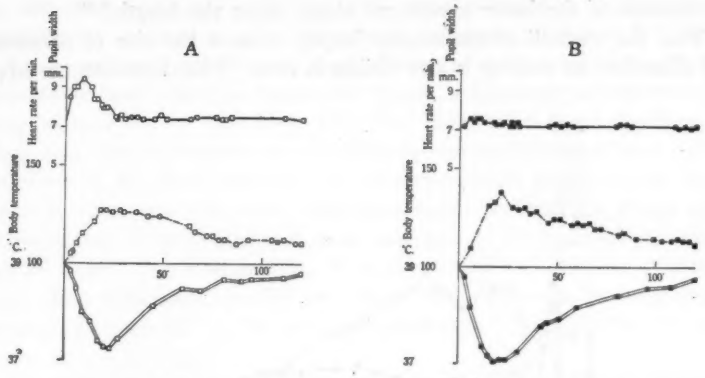

FIG. 27. Paradoxical pupil dilatation and cardio-acceleration on introduction of cold water in dogs through the stomach fistula. (Nagakura)

Pupil width in solid line; heart rate chain line, and anal temperature double line.

Light quadrates show the estimation in normal dogs and solid quadrates those in the medulli-adrenalectomized.

wholly justifiable to speak of the total abolishment of the paradox.

Generally speaking, the paradoxical pupil dilatation on cooling an animal sets in much earlier and fades out much earlier than that of adrenaline oversecretion and hyperglycemia, which almost simultaneously take place. Latency

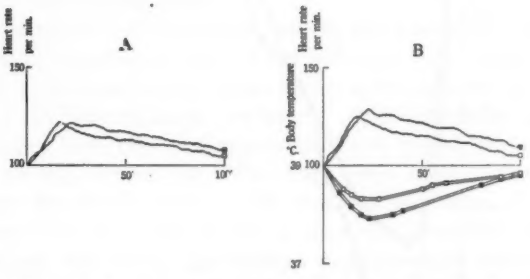

FIG. 28. A, B. Introduction of cold water through the gastric fistula and rate of denervated heart and anal temperature. (Wada)

Normal cases (6 in number) are indicated with light quadrates and medulli-adrenalectomized ones of the same number with solid quadrates.

The heart rate was noted every 5 minutes, which resulted in the figures given.

(A) The heart rates are recalculated on the body temperature at that time, as is usual in this book.

(B) The heart rates are treated in their actual values.

and duration of the latter events are about twice the length.[3,10]

That the medulli-adrenalectomy largely reduces the size of paradoxical pupil dilatation on cooling is also visible in cats. The dilatation on dipping

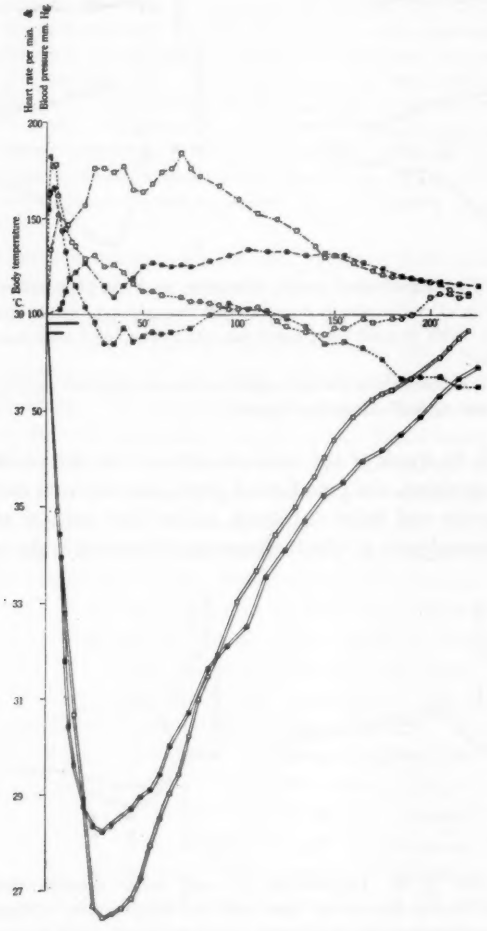

FIG. 29. Denervated heart rate of dogs in the cold bath experiments. (Wada & Fuzii)

Denervated heart rate is shown with chain line, arterial blood pressure dotted line, and anal temperature double line. Mean values in two normal dogs are indicated with light quadrates, and those in two medulli-adrenalectomized ones with solid quadrates.

in a cold bath is 3 mm. in the cat with intact adrenals. Double medulli-adrenalectomy or double splanchnicotomy reduces it to 0.8 to 0.7 mm.[11]

In the cooling experiment in dogs, where the denervated heart was tested at the same time with the denervated pupil, a fundamental difference was clearly worked out.[10] (See Fig. 27) The denervated heart accelerates on cooling also, but the latency and duration of tachycardia were very different from those of the pupil reaction, i.e. incomparatively longer in the former. Here cold water was introduced into the stomach through the gastric fistula. The maximum of tachycardia in each case was 10 to 40 beats per minute, mean 20 beats; the latency 5 to 10 minutes, and the duration one to two hours. The demedullation did not affect the tachycardia in magnitude of duration: 25 beats (20 to 30) increased on cooling. Tachycardia was visible for one and a half-hours.

The cold water introduction into the stomach of dogs with the completely denervated heart is a repetition of the previous work of Wada.[11] He conducted the cold water introduction before and after the demedullation of adrenals. The cardiac acceleration and shiverings of quite the same degree and duration were noted in both kinds of experiments. The amount of heat-debt in both the experiments of Wada and of Nagakura was taken somewhat greater than Cannon. Ruling out the medulli-adrenal function did not change at all the cardiac acceleration due to cooling, contrary to the latter investigator. (Fig. 28 A & B) That the cardio-acceleration was definitely greater, though insignificantly, in the medulli-adrenalectomized dog is justifiably accounted for by a greater fall in the body temperature. This increases a further secretion of sympathins.

In contrast to the cooling experiment by the introduction of cold water into the stomach, dipping of dogs in the ice-water bath, which results in a big fall of the body temperature, gives also a quite different outcome re the heart rate and the arterial blood pressure.[13] The demedullation of adrenals distinctly reduces here the cardioacceleration and blood pressure elevation, as illustrated in Figs 29 and 30. The dogs, given in the former figure had the completely denervated heart,[13] and those in the latter the intactly innervated heart.[1] In the latter group of dogs the adrenaline hydrochloride solution injection was run in our manner simultaneously with the cold application. The outcome re cardio-acceleration in the latter set of experiment well coincides with that in the normal dog, produced by cold bathing alone.

The pulse rate given in this and the next chapter is recalculated, the effect of body temperature being taken into account in the manner described on p. 142. The relation between cooling, adrenaline secretion and heart rate is also discussed in detail in Chapter VIII, on pp. 143–150.

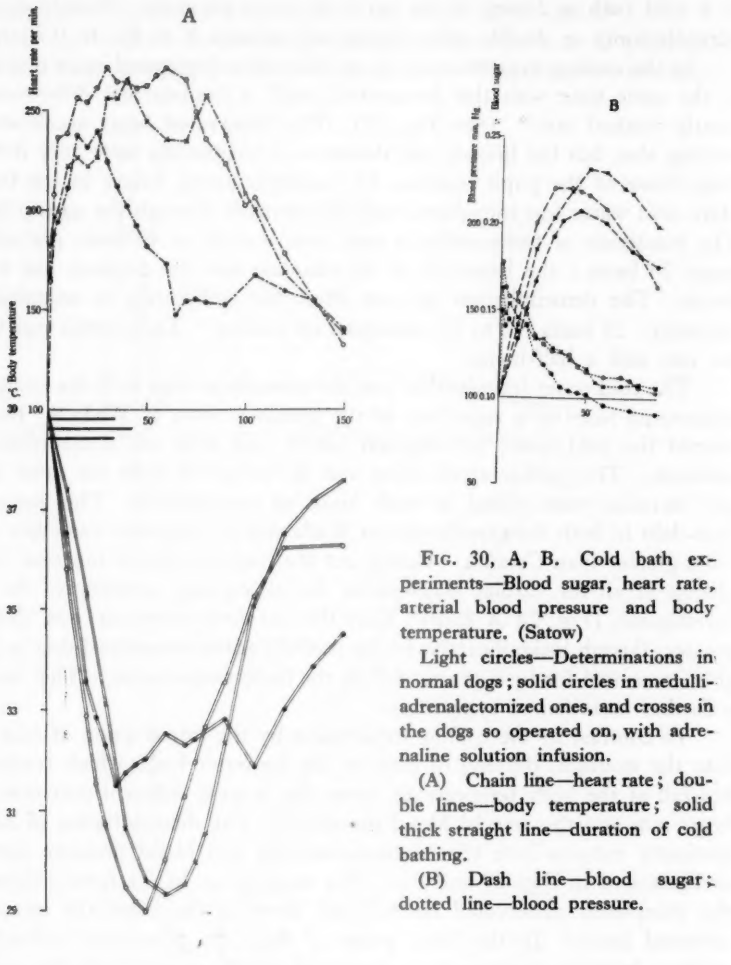

FIG. 30, A, B. Cold bath experiments—Blood sugar, heart rate, arterial blood pressure and body temperature. (Satow)

Light circles—Determinations in normal dogs; solid circles in medulli-adrenalectomized ones, and crosses in the dogs so operated on, with adrenaline solution infusion.

(A) Chain line—heart rate; double lines—body temperature; solid thick straight line—duration of cold bathing.

(B) Dash line—blood sugar; dotted line—blood pressure.

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8) *Id.*, *Ibid.*, 78.

9) Cannon, *Am. J. Physiol.*, 1898, **1**, p. 363; Cannon, Shohl and Wright, *Ibid.*, 1911 (-12), **29**, 282.
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12) Wada, *Ibid.*, 1935, **26**, 546.
13) Wada and Fuzii, *Ibid.*, 1940, **37**, 505.

Heating

When the whole body surface of a normal dog is heated, for example by means of a rubber coat filled with hot water, in such a scale as to elevate the anal temperature over 41°C., first the adrenaline secretion augments definitely. Fig. 31 shows it.[1] Heating was applied for 10 to 15 minutes. The highest rate of mean 0.3 γ (0.1 to 0.8 γ) per kg.-min. was reached 10 minutes after the start of heating. The oversecretion lasts for about one hour, and the whole oversecretion 6 γ per kg. in 50 minutes. The shape of oversecretion of adrenaline on heating might be taken as Type B_1.

The heat-puncture commonly does not effect any increase in the blood sugar concentration in rabbits, besides a transitory hyperglycemia due to mechanical seizure.[2]

When a heat-punctured rabbit is placed in a heated room and the anal temperature reaches 40° to 42°C. or higher, hyperglycemia of 0.2% (0.14–0.28%) takes place. Double splanchnicotomy and double adrenalectomy as well largely reduce the hyperglycemia in rabbits, heat-punctured and placed in environmental temperature of 50°C.[3] But the latter be raised to 60°C. the blood sugar in the rabbit so operated on reaches 0.14% in contrast to 0.16% in the control animal, viz. the rabbit with normal adrenal function but heat-punctured.[4]

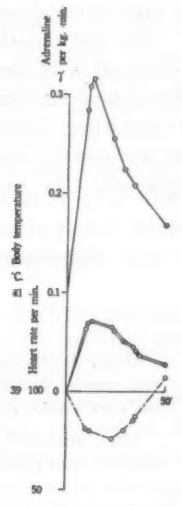

Fig. 31. Application of heat upon the body surface in normal dogs, and the adrenaline secretion. (S. Saito)
Adrenaline secretion rate in solid line, pulse rate in chain line and anal temperature in double line.

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Narcotics, Hypnotics

General narcotics inhibit the adrenal secretion from the adrenals, a fact worthy of especial note, but they invariably act hyperglycemic. Ether and chloroform are good examples of this. Most agencies which increase the blood sugar concentration in organisms also act to accelerate the adrenaline secretion as a rule, in contrast to the narcotics. Hypnotics have either a little action upon both adrenaline secretion and blood sugar content, or none at all upon them both. Avertin, evipan and veronal are examples. We have no experience about the effects of alcohol.

General anesthetics such as ether, chloroform, invariably bring about a diminution of the adrenaline secretion rate, chiefly or solely by that of the adrenaline concentration in the blood from the adrenals. If they are applied to cause a deep anesthesia such as to permit the carrying out of major operations, the preliminary rate is reduced by thirty to sixty per cent;[1,2,3] schematically expressed. For example:

TABLE XII

Influence of Anesthesia upon Adrenaline Secretion Rate in Dogs

	Adrenaline secretion rate (per kg.-min.)			
	Preliminary (1)	Under ether (2)	Reduction (1–2)	Rate of reduction 1 : (1–2)
Dogs, de-afferented, non-narcotized, provided with the cava pocket preparation	0.64 γ	0.25 γ	0.4 γ	60%
Dogs, etherized, provided with the cava pocket preparation	1.2 γ (estimated ten minutes after stopping etherization)	0.5 γ	0.7 γ	60%

It is superfluous to add here that the deeper the narcosis the smaller the adrenaline secretion; in reality the Cleveland physiologists take the "spontaneous liberation rate" as 0.2 γ per kg.-min.[4] That is to say, ether produces there eighty per cent reduction in the adrenaline secretion rate.

Such a deepness of anesthesia results in ineffectiveness of sensory stimulation, asphyxia, etc. in accelerating the adrenaline secretion. This ineffectiveness effects in turn a remarkable constancy in the "spontaneous liberation of adrenaline" in their hands. And this constancy leads them in turn to believe in, reasonably but in fact paradoxically, the correctness of their method for determining variations in the adrenaline secretion. Here it may be added incidentally, that in their later stage of researches Stewart and Rogoff called the method the intestinal method, but it is correct, however, to term it the cava pocket method, or more exactly, the cava pocket method under anesthesia. It is immaterial there whether the adrenaline in the adrenal venous blood is

determined by means of the rabbit intestine strip or any other method, biologically or chemically.

With the anesthesia deep enough to cause the so-called " spontaneous liberation " of the Cleveland physiologists, i.e. 0.2 γ per kg.-min., the secretion rate is almost or wholly the same regardless of whether the blood is collected through the lumbar route or through the cava pocket, as shown by them[5] and ourselves[6] as well. But with the anesthesia less deep, the latter method yields a much larger value than the former, as 0.6 γ per kg.-min. against 0.2 γ.[6] Preparing the cava pocket method is a more major operation to the organism than preparing the lumbar route method.

The fact that definitely different degrees of the so-called " spontaneous liberation " as 0.8 γ per kg.-min., 0.5 γ[3] or 0.2 γ[4] were obtained, notwithstanding that one and the same procedures, i.e. combination of the cava pocket method and the intestinal segment method were used, can be justifiably explained on the ground of different degrees of anesthesia.

Samples taken 30 seconds after its commencement already show the diminishing effect of etherization, and when rested ten minutes after discontinuation of etherization, the reduction is found either more deepened, lessened or already abolished. It sometimes happens that a larger rate than the preliminary is present.

Thus the depressing effect of surgical anesthetics upon the adrenaline secretion is very powerful in the following cases: asphyxia,[7] sensory stimulation,[8,3] shock by peptone,[9] or hemorrhage,[10] piqûre diabétique.[11]

On the other hand there are some other agencies, the accelerating action of which upon the adrenaline secretion is weakened by ether, chloroform, etc., but not so profoundly as by the agencies given above. They are pilocarpine,[12] strychnine,[13] morphine,[14] etc. And atropine[15] and β-tetra[16] leave the hyperadrenalinemic ability almost or entirely unaffected by surgical anesthetics; further, physostigmine shows seemingly a more powerful hyperadrenalinemic effect in the narcotized animal.[17]

Aliphatic hypnotics commonly act to reduce the adrenaline secretion, though on a far smaller scale compared with ether, chloroform, or exert no influence thereupon. They are chloral hydrate,[18] chloralose,[19] avertin[20] evipan,[21] veronal.[22] They were proven to act in that way either by testing their influence upon the basal secretion of adrenaline in the dog, de-afferented and provided with the lumbar route preparation, or by administering an accelerating agent in the dog under a given hypnotic. Depressing action of the three latter hypnotics is especially weak; the hyperadrenalinemic action of insulin[23,24] and morphine[25] is not influenced by them.

Aliphatic hypnotics might thus be recommended to be applied widely in the study of adrenaline secretion.

Among the anesthetics tested here, urethane is the only one with an

accelerating action on the adrenaline secretion. It was given in cats, with the cava pocket preparation, intravenously only, but in dogs de-afferented, intravenously or subcutaneously. Urethane of 1–3 g. per kg. produces in dogs a small hyperadrenalinemia of 3–4 times the normal rate. The acme was reached one to two hours after i.v. injection, and two to three hours after hypodermical application. The secretion is of Type C. The total amount of secretion: i.v. 12 γ per kg. (170 minutes) and subc. 20 γ (265 minutes).[26]

The effect of ether and chloroform is always hyperglycemic, whereas they always reduce the adrenaline secretion, as proven above. So far as I know there is no evidence that ether and similar anesthetics act to reduce the hyperglycemia elicitable by other agencies.

Degree of hyperglycemia in rabbits, for example, by ether, depends upon the depth of narcosis: 0.15 to 0.25% in shallow narcosis, and 0.25 to 0.45% in deep.[27] In dogs 0.2% or more hyperglycemia is common under ether.[28,29] Here the adrenaline secretion is diminishing.[29]

Protection against the body temperature fall diminishes the blood sugar elevation due to ether in dogs,[28] contrary to the case with rabbits.[11] Double splanchnicotomy[27] or adrenalectomy diminishes it too, as from 0.2–0.25% to 0.15%, roughly speaking. When the ether anesthesia is deep enough, the effect of double adrenalectomy upon the magnitude of hyperglycemia appears very pronounced.[31]

Urethane, which is only hyperadrenalinemic in action, acts similarly to the volatile narcotics in respect to the glycemic action.[11] Giving 1 to 2 g. urethane orally or subcutaneously to rabbits produces a hyperglycemia with a wavelike fluctuation of intensity, the duration of one wave lasting for some hours. Double splanchnicotomy has no effect thereupon at all,[32] but double adrenalectomy reduces the urethane hyperglycemia, as from 0.2% to 0..15%.[33]

Protection against a body temperature fall acts to diminish the grade of hyperglycemia in rabbits by urethane.[32] Whether or not the protection against the body temperature fall, can depress the blood sugar increase causable by anesthetics depends upon the depth of anesthesia in general, the several data given above there and here be taken collectively.[27,32]

Hyperglycemia in rabbits by chloretone is also reduced by double splanchnicotomy or adrenalectomy, 0.12% being yielded in those animals operated on against the 0.2% in normal ones.[34]

Contrary to surgical anesthetics, hypnotics such as avertin[20] and veronal[22] which have little or no effect upon the adrenaline secretion mechanism have also no influence upon the carbohydrate metabolism or, if any, only a light depressing effect.

Finally there is an exception re the adrenalinemic and glycemic action of anesthetics. Chloralose has a tendency to decrease the adrenaline secretion by virtue of a reduction of blood flow through the adrenals. And it reduces the blood sugar concentration to about 0.07–0.08%.[11]

The fact that the Cleveland physiologists always obtained a remarkably large hyperglycemia, such as 0.3–0.4%, in the doubly adrenalectomized rabbits on giving ether and others[35] will be referred to in the next chapter.

Narcotics such as ether, chloroform, urethane, etc. diminish the adrenaline load of glands, unless protected by excluding the innervation.*

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8) *Id.*, *Ibid.*, 1923, **4**, 166; 1924, **4**, 465.
9) M. Watanabe, *Ibid.*, 1927, **9**, 412.
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15) Stewart and Rogoff, *J. Pharm. Exp. Ther.*, 1920, **16**, 71; H. Sato, Hatano and Muto, *Tohoku J. Exp. Med.*, 1938, **34**, 289. (By comparison)
16) Sugawara, *Tohoku J. Exp. Med.*, 1927, **9**, 368 (cats); H. Sato and Sugawara, *Ibid.*, 1930, **16**, 580 (dogs).
17) Stewart and Rogoff, *J. Pharm. Exp. Ther.*, 1921, **17**, 227 (cats); F. Watanabe, *Tohoku J. Exp. Med.*, 1935, **27**, 404 (dogs). (By comparison)

Remarks: The comparisons here made are based not only on the data from different conditions of experimentation, but also on various kinds of animals (cats and dogs). They can not be taken therefore as rigidly accurate.

18) Kodama, *Tohoku J. Exp. Med.*, 1924, **5**, 157 (cats).
19) H. Sato and Ohmi, *Ibid.*, 1933, **21**, 433.
20) Sato, Satow and Degti, *Ibid.*, 1934, **24**, 485.
21) Tatsuzi Suzuki, K. Saito and Kobayasi, *Ibid.*, 1951, **54**, 261.
22) H. Sato and Degti, *Ibid.*, 1935, **25**, 115.
23) H. Sato, Satow and Degti, *Ibid.*, 1934, **24**, 495 (insulin-avertin).
24) Tatsuzi Suzuki and K. Saito, *Ibid.*, 1951, **54**, 309 (insulin-evipan).
25) H. Sato, Degti and Satow, *Ibid.*, 1935, **25**, 107 (morphine-avertin).
26) Kodama, *Ibid.*, 1930, **15**, 11; H. Sato and Aomura, *Ibid.*, 1930, **15**, 17.
27) Fujii, *Ibid.*, 1921, **2**, 169 (rabbits, splanchnicotomy).
28) Tachi, Takai and Fujii, *Ibid.*, 1926, **7**, 411.
29) Kodama, *Ibid.*, 1924, **4**, 601.
30) Tachi and Hirayama, *Ibid.*, 1926, **8**, 41 (rabbits, adrenalectomy).
31) Ohmi, *Ibid.*, 1933, **21**, 116 (rabbits).
32) Hirayama, *Ibid.*, 1926, **7**, 364 (rabbits).

33) Tachi and Hirayama, *Ibid*, 1928, **10**, 191 (rabbits).

34) Tada, *Ibid.*, 1931, **17**, 574 (rabbits).

35) Stewart and Rogoff, *Am. J. Physiol.*, 1918, **46**, 90 (piqûre, asphyxia); *Ibid.*, 1920, **51**, 366 (ether, asphyxia); *Ibid.*, 1922, **62**, 93 (morphine).

• Unfortunately the data of Elliott, J. Physiol, 1912, **44**, pp. 388–390 are cited erroneously in the valuable monography of Hartman and Bromwell, *The adrenal gland*, Philadelphia 1949, 131.

Adrenaline

Adrenaline itself exerts neither an accelerating influence upon the adrenaline secretion rate[1] nor a diminishing influence upon the adrenaline content of the adrenals.[2] If there exists already some excess secretion before administration of adrenaline, the adrenaline exercises a sedative influence thereupon.[1]

The statemaent is induced from the experiment on dogs, almost normal where blood samples were collected simultaneously from the adrenal vein, any artery and vein as well. The intravenous dosage was 0.06 to 0.3 mg. adrenaline hydrochloride per kg. body weight.[1]

A diminution in the flow rate through the adrenal vein in dogs under narcosis was recorded by Brauch, Brown and Rein under their experimental conditions when adrenaline was given in a dose of 1/10 000–1/500 mg. per kg.[3]

References

1) Sugawara, S. Saito and Nemoto, *Tohoku J. Exp. Med.*, 1926, **9**, 149.

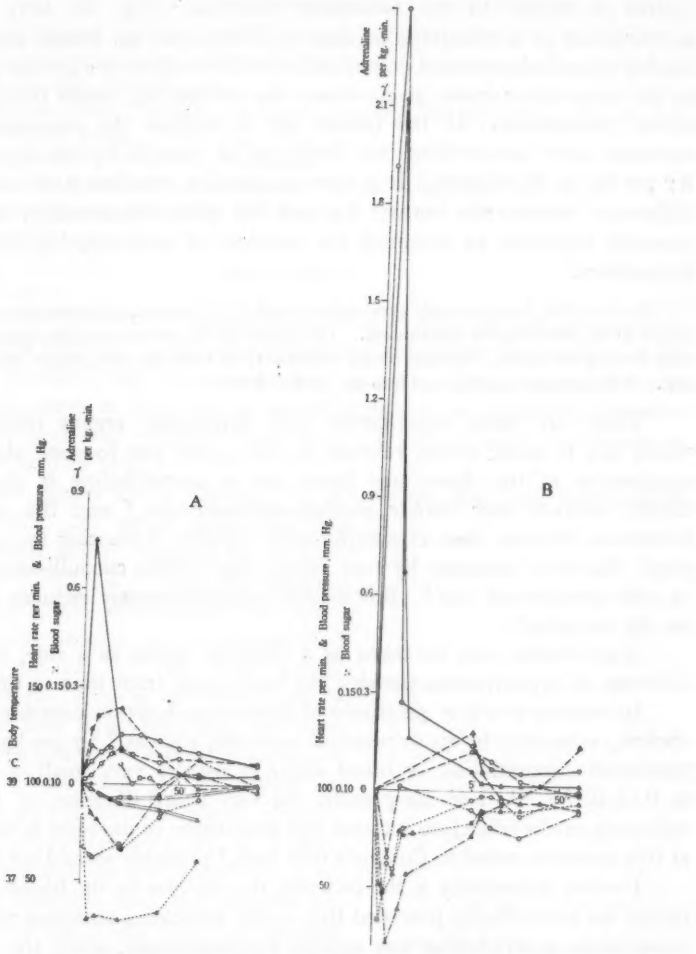
2) Elliott, *J. Physiol.*, 1912, **44**, p. 393 (cats, unilateral splanchnicotomy, etherization; pressoric action estimated.); Zy. Kanowaka, *Tohoku J. Exp. Med.*, 1934, **24**, 572 (rabbits, Suto-Kojima's method.); etc.

3) Brauch, Brown and Rein, *Ztschr. f. Biol.*, 1935, **96**, 42.

Acetylcholine

Acetylcholine produces a rapid increase in the adrenaline secretion rate in the normal dog under our experimental conditions. On an intravenous application of 5 mg. per kg. the rate increases to 2.5 γ per kg.-min. on an average, 50 times acceleration, *ad maximum* 3.5 γ. But the accelerating period is quite brief; two minutes after injection the rate is almost back to the pre-existing rate. The total excess amount of adrenaline is therefore only 2 γ per kg. body weight. The splanchnicotomy does not so much interfere with the accelerating power of acetylcholine, the averages being 2 γ per kg.-min. and 1.3 γ per kg. Type A_2, approximately said, but strictly A_1.[1] (Fig. 32, right)

Hypodermically applied, acetylcholine however yields somewhat different



FIG, 32, A, B. Influence of acetylcholine upon adrenaline secretion, etc. "A" shows those produced by subcutaneous injection of 15 mg. per kg. (E. Inaba), and "B" those by i.v. injection of 5 mg. per kg. (Wada et al.)

The adrenaline secretion rate scale is one-third of that commonly applied in this book, in both figures, i.e. "A" & "B". The time scale for the first 5 minutes in "B" is enlarged tenfold.

The values in normal cases are shown with light circles and those in splanchnicotomized with light triangles.

Adrenaline secretion rate—solid line, blood sugar content—broken line, arterial pressure—dotted line, pulse rate—chain line, anal temperature—double line.

figures in respect to the adrenaline secretion.[2] (Fig. 32, left) Whereas acetylcholine in a subcutaneous dose of 15 mg. per kg. brings about in the doubly splanchnicotomized dog a similar but slow curve of a similar magnitude to the intravenous cases, given above, the normal dog reacts there with still slower fluctuations. In the former set of animals the maximum rate of secretion after acetylcholine was computed as mean 0.7 γ per kg.-min., and 8 γ per kg. in 90 minutes. It is open to question, whether such a remarkable difference between the normal dog and the splanchnicotomized one can be correctly explained by assuming the existence of inhibiting-like fibres in the splanchnics.

On the other hand, a small acceleration, such as 2–8 times, of adrenaline was reported of cats in the acetylcholine intoxication. This figure should not however be compared directly with those given above. Adrenal bloods were collected from the cava pocket prepared under dial. The hormone was injected into the coeliac artery.[3]

There are some experiments with denervated organs (heart, pupil), which are to some extent relevant to the query put forward above. The acceleration of the denervated heart due to acetylcholine in dogs is distinctly inferior after double medulli-adrenalectomy,[4] and the intravenous minimum effective dose of acetylcholine capable of eliciting the paradoxical pupil dilatation increases by four times after double medulli-adrenalectomy in non-anesthetized cats.[5] But double splanchnicotomy reduces it by half, on the contrary.[5]

Acetylcholine can be taken as a glycemic agent as a rule, though occurrence of hypoglycemia thereby has been noted from time to time.[6]

In contrast to a very great rate of adrenaline secretion causable by acetylcholine, especially by its intravenous injection, such as 2.5 γ per kg.-min., the maximum concentration of blood sugar is usually very small in dogs, such as 0.12–0.13%.[2,1] For the paradox the very short duration of hyperadrenalinemia can be taken into account, but stimulation of the islets in the pancreas at this occasion, noted in Cushny's text book,[7] probably should not be ignored.

Further noteworthy is the fact that the increase in the blood sugar concentration undoubtedly preceded that in the adrenaline secretion rate in some cases where acetylcholine was applied subcutaneously, while the splanchnic nerves remained untouched.[2] This gives evidence that the oversecretion of adrenaline is not the sole cause of increase in the blood sugar here, or otherwise expressed, that acetylcholine is capable of increasing the blood sugar without relying on the adrenaline action.

Acetylcholine produces a reduction of the adrenaline content of glands in rabbits, irrespective of whether the innervation remains intact or not; the recovery too.[8]

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2) E. Inaba, *Tohoku J. Exp. Med.*, 1935, **27**, 245.
3) Taneiti, *Ibid.*, 1940, **38**, 147.
4) Youmans, Haney, Lindgren and Kastens, *Proc. Soc. Exp. Biol. Med.*, 1941, **47**, 247.
5) S. Kojima, *Tohoku J. Exp. Med.*, 1942, **42**, 72.
6) Onti, *Ibid.*, 1941, **41**, 102 (Literatures).
7) Cushny's *Pharmacology & thereapeutics*, 12. ed., Philadelphia 1940, 488.
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PILOCARPINE

Pilocarpine produces a hyperadrenalinemia in both the non-anesthetized dog, provided with the lumber route preparation[1] and the dog and cat under anesthesia, provided with the cava pocket,[2] as well. The accelerating action is almost the same in both sets of experiments, i.e. no significant influence of anesthsia upon the effective doses of the drug, or upon the degree of acceleration is to be seen.

Numeral figures are as follows: in the normal dog, non-anesthetized, 1, 2 and 4 mg. per kg. of pilocarpine was given hypodermically, whereby an acceleration of 0.03 γ, 0.2 γ and 0.3 γ per kg.-min., i.e. 2, 5 & 8 times acceleration took place, with the total amount of oversecretion of 2,5 γ, 10 γ and 20 γ per kg. body weight. The time when the peak was noted may be taken as one hour after injection, the whole duration of acceleration exceeding two hours.

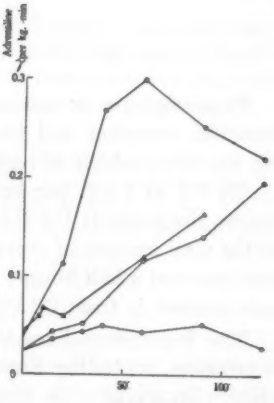

FIG. 33. Pilocarpine, subcutaneously injected, upon adrenaline secretion. (F. Watanabe)

On normal dogs (light circles): from upper 4 mg. per kg., 2 mg. & 1 mg. On a splanchnicotomized one 2 mg. per kg. was given (light triangles).

Giving 1 and 2 mg. per kg. pilocarpine produced in an anesthetized dog an acceleration in the adrenaline secretion of 2 and 3 times. Depending on the magnitude of the pre-existing secretion rate, the absolute rate of acceleration was 1 and 2.5 γ per kg.-min.

The minimum effective dose for causing the hyperadrenalinemia is 1 mg. per kg., irrespective of whether the animal be non-anesthetized or anesthetized, or whether it be intravenously or subcutaneously applied.

Splanchnicotomy reduces largely the adrenaline oversecretion due to pilocarpine, but never annuls.[1]

The blood sugar concentration varies in the normal dog in a similar

manner to the adrenaline secretion. Giving 1, 2 and 4 mg. pilocarpine per kg. increases the former to 0.15%, 0.16% and 0.175%; double splanchnicotomy reduces the hyperglycemia there.[1] Double splanchnicotomy and double adrenalectomy diminish the intensity of pilocarpine hyperglycemia in rabbits from 0.15% to 0.13%.[3]

Pulse rate is also accelerated by pilocarpine, hypodermically given, and especially in the early stage and lasts for a half to one hour in the normal dog. And while the adrenaline secretion and blood sugar are increasing further, the heart begins to beat slow, so that one hour after the injection it is beating at a smaller rate than the pre-existing. Double splanchnicotomy acts mostly to depress the cardiac acceleration and the body temperature elevation.[1]

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1) F. Watanabe, *Tohoku J. Exp. Med.*, 1935, **27**, 390.

2) Hirano, *Ibid.*, 1939, **37**, 119.

3) Tada, *Ibid.*, 1933, **20**, 275. (The experimental data of Viale and Napoleoni are quoted there on p. 282, plainly but with critical connotation. Cf. Sollmann, *A manual of pharmacology*, 7. ed., 1948 Philadelphia & London, 327.)

PHYSOSTIGMINE

Physostigmine or eserine in the unanesthetized dog acts to increase the adrenaline secretion and blood sugar.[1] The increasing and decreasing of both are slow, which is very similar to the effect of pilocarpine.

On 0.5 to 1 mg. per kg. physostigmine, the greatest acceleration of adrenaline discharge is 0.1–0.15 γ per kg.-min., i.e. 2 to 8 times acceleration, and the total amount of oversecretion 5 γ per kg. The whole duration covers about two and a half hours; the type of course is C. The acme of the blood sugar content is from 0.13% to 0.15%.

The hyperadrenalinemic action of this drug has been proved by all the investigators, including Stewart and Rogoff who were unable to find it on testing pilocarpine; 14 times acceleration on subcutaneous application of 1 mg. the drug per kg. was the greatest acceleration there.[2]

Whether the adrenal venous blood is collected by means of the cava pocket under anesthesia or through the lumbar route without anesthesia has no bearing upon the degree of acceleration causable by physostigmine, just as in the case of pilocarpine. The absolute rate depends here upon the pre-existing value, as pointed out there too.[3]

As to the question of how double splanchnicotomy alters the adrenalinemic ability of physostigmine, there are divergent opinions: a majority of investigators obtained a positive outcome even with the depriving of the

splanchnic nerves,[4,3] contrary to Stewart and Rogoff.[2] In the former sets of experiment the neurotomy was done either acutely or previously.

Physostigmine enhances the accelerating action of KCl upon the adrenaline secretion in the normal dog, roughly by three times.[5]

Double splanchnicotomy and especially double adrenalectomy lessen the increase of the blood sugar due to physostigmine in rabbits.[6] The drug diminishes the adrenaline content of glands in rabbits.[6]

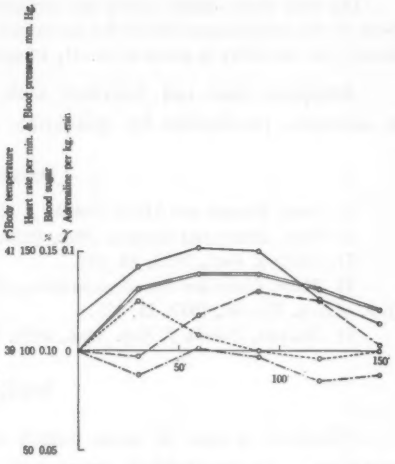

FIG. 34. Physostigmine in normal dogs, subcutaneously given in 0.5 mg. per kg. (F. Watanabe)

Adrenaline secretion rate—solid line, blood sugar—broken line, arterial pressure —dotted line, pulse rate—chain line, anal temperature—double line.

REFERENCES

1) F. Watanabe, *Tohoku J. Exp. Med.*, 1935, **27**, 404.

2) Stewart and Rogoff, *J. Pharm. Exp. Ther.*, 1921, **17**, 227.

3) Suzuki, *Tohoku J. Exp. Med.*, 1950, **52**, 385.

4) Tscheboksaroff, *Pflüger Arch.*, 1911, **137**, 96–98; Houssay and Molinelli, *C.r. Soc. Biol.*, 1925, **93**, 1133; *Am. J. Physiol.*, 1926, **77**, 184; Tournade and Chevillot, *C.r. Soc. Biol.*, 1937, **124**, 565; Satow, *Tohoku J. Exp. Med.*, 1938, **32**, 257.

5) Suzuki, Matumoto, Sasaki and Nunokawa, *Tohoku J. Exp. Med.*, 1951, **54**, 253.

6) Tada, *Ibid.*, 1933, **20**, 275.

ATROPINE

In the normal dog 5–10 mg. per kg. atropine, intravenously given, soon produces a small elevation in the adrenaline secretion rate, say by two to three times. The hyperadrenalinemia lasts for one hour or longer, and the blood sugar concentration increases in parallel with it.[1]

The drug does not interfere with the hyperadrenalinemia causable by morphine or insulin. If combined with the latter the increase in the adrenaline secretion rate is greater than that producible by the latter alone.[2]

Although it has been said that atropine causes an evident increase in the adrenaline content in the adrenals if the amount be sufficiently large, we have never been able to obtain such evidence, notwithstanding such enormous amounts of the drug as 70 mg. per kg. were given and then the rabbits were killed after 5 minutes, 60 minutes or 6 hours (Here 135 mg. atropine were injected in 11 times).[3]

The data above clearly prove the invalidity of the idea that only the parasympathetic fibres in the splanchnics control the adrenaline secretion.[4] Another experiment that proves directly the invalidity is given on p. 41, Chapter VI.

Atropine does not interfere with the reduction of adrenaline content in adrenals, producible by splanchnic stimulation, bleeding or insulin.[5]

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1) Sato, Hatano and Muto, *Tohoku J. Exp. Med.*, 1938, **34**, 289.
2) Sato, Muto and Hatano, *Ibid.*, 1938, **34**, 393.
3) Sazawa, *Ibid.*, 1938, **34**, 277.
4) Kuré, *Ueber den Spinalsympathikus*, 1931 Basel, 48 ff.; Kuré, Y. Wada and Okinaka, *Q. J. of Exp. Physiol.*, 1932, **21**, 227.
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Nicotine

Nicotine is one of most potent agents for accelerating the adrenaline secretion. An enormously great rate of secretion, such as 20 γ adrenaline per kg.-min., i.e. several hundred times the pre-existing value, was demonstrated in the normal dog, on i.v. administration of 1 mg. nicotine per kg. body weight. The peak, mean 8 γ adrenaline per kg.-min. (the greatest 20 γ), becomes manifest 15 seconds after injection, and one minute after injection the secretion has half the value of the maximum. Five minutes after the injection about ten times the basal rate is to be found, and thirty minutes are needed to see a complete recovery. Nicotine thus accelerates the adrenaline secretion remarkably quickly and profusely on the one hand, but the rate also goes down quickly. The total amount of oversecretion is computed as mean 20 γ per kg. The nicotine hyperadrenalinemia is of Type A_2.[1]

In this experiment of Wada et al. the adrenal blood was collected successively from the start of the injection each with 30-second period for two minutes; then with longer intervals successive collections were made. If ten-second collections be adopted a more natural course would be realized.

If this period of the greatest acceleration be missed when collecting samples, or otherwise expressed, if the collection be commenced five minutes after nicotine, the secretion curve for nicotine injection would be modified as shown in the next figure (Fig. 35, B), a surprising contrast to Fig. 35, A. (It may be noticed here especially: the scale of adrenaline secretion rate is on the left figure taken as one tenth of the common scale in this book, e.g. that on the right figure, and the time scale on the former is taken for first-five minutes as ten times as the common. Extraordinary acceleration and rapidity of variation compel us to modify the scales thus. Two low curves with thick solid lines on the right figure are drawn with the same scales on the left side figure for the sake of comparison.)

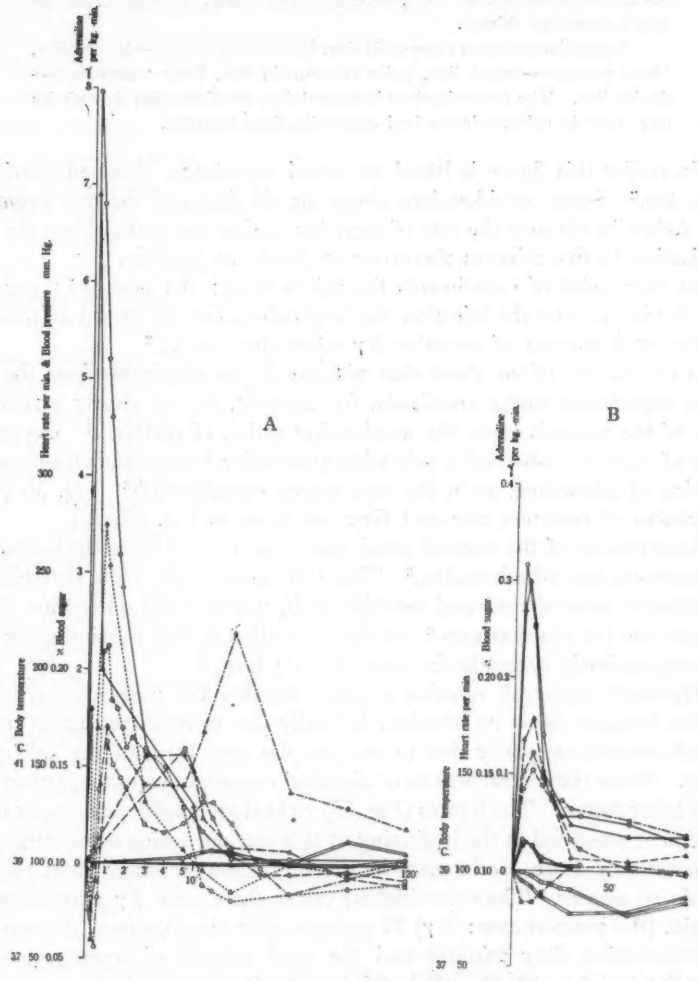

FIG. 35, A, B. Adrenaline secretion rate, blood sugar concentration, etc. on 1 mg. per kg. nicotine, i.v. given.

In the experiments shown in " B " the determinations were started first 5 minutes after the injection (F. Watanabe); in those in " A " collection of adrenal blood was started simultaneously with the injection of nicotine solution and carried out in short intervals (Wada et al.).

Adrenaline secretion rate scale is given in " A " with one-tenth of that commonly applied in this book, because of its being too large. In " B " it is given in two different ways: once with the common scale and once with

the same scale as that in " A ", for comparison's sake; here solid lines are drawn somewhat thicker.

Adrenaline secretion rate—solid line, blood sugar content—broken line, blood pressure—dotted line, pulse rate—chain line, body temperature—double line. The determinations in normal dogs are given with light circles and those in splanchnicotomized ones with light triangles.

In reality this figure is based on actual experience, obtained previously by us, too.[2] Some considerations about the old data and the new prompted us to follow in advance the rate of secretion during the period from the start of injection to five minutes thereafter as closely as possible.

In that series of experiments the values were: the peak 0.3 γ per kg.-min., 5 minutes after the injection, the hyperadrenalinemic period 25 minutes, and the total amount of secretion 3 γ adrenaline per kg.[2]

A comparison of the above data with the figures obtainable from the cava pocket experiment under anesthesia, for example, proves clearly a reducing effect of the narcosis upon the accelerating action of nictoine.[1] Depressive action of nicotine is observable only when there exists beforehand an augmented secretion of adrenaline, as in the cava pocket experiment.[4,3] Fig. 36 shows it. (Scales of secretion rate and time are same as Fig. 35, A.)

Denervation of the adrenal gland cannot so modify there the increase in the secretion rate of adrenaline.[3] This fact induced the view that nicotine acts directly upon the adrenal medulla itself, whereby the adrenaline is discharged into the blood stream,[3] and the adrenaline is thus nothing other than the postganglionic sympathetic fibres in fluid form.[5]

However there still remains a query whether the oversecretion of adrenaline brought about by nicotine, is totally due to its direct action on the adrenal medulla or partly due to the impulse sent through the splanchnic nerves. From the central actions of nicotine[6] one might expect a participation of the latter factor. The figures (Fig. 35) yielded after splanchnicotomy in the experiment presented at the beginning of this section, which demonstrate first an enormously large acceleration in the adrenaline secretion could possibly provide an answer. The mean highest values there were 7 γ adrenaline per kg.-min. (the greatest case: 9 γ) 15 seconds after the injection, the duration of oversecretion sixty minutes and the total amount of oversecretion 16 γ adrenaline per kg. On the whole this acceleration seems a little smaller than the case with the intact innervation.

In two dogs, nicotine was given first when the splanchnic nerves were still sound, and later after the neurotomy was done. In one dog the secretion rate in the first 30-seconds period beginning with the start of nicotine injection in a dose of 1 mg. per kg. was 10 γ adrenaline per kg.-min. from one gland, 500 times acceleration, and in the next 30-seconds period 6.7 γ per kg.-min., while the gland was still intactly innervated; the denervation of

the gland reduced them to 4.5 γ per kg.-min. (225 times acceleration) and 1.4 γ respectively. The figures in the other dog were respectively 7 γ adrenaline per kg.-min. (300 times), and 4.5 γ before the denervation, and 2.4 γ per kg.-min. (240 times) and 0.6 γ.[1]

The outcome on one and the same dog, applying to each of the two dogs subjected twice to nicotine, permit us acknowledge that denervating the adrenal gland has a reducing effect on the hyperadrenalinemic activity of nicotine. In order to evaluate with some definiteness the role of each factor, peripheral and central, some experiments similar to the above examples to be added. Preferably splanchnic nerves should be cut a sufficient time in advance. In similar experiments on cats provided with the cava pocket under anesthesia, the nicotine hyperadrenalinemia after double splanchnicotomy was definitely larger when the drug was applied several days after neurotomy than when it was done soon thereafter.[3,*]

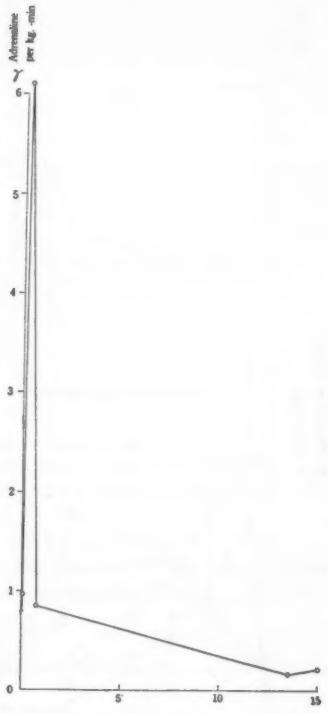

FIG. 36. Adrenaline secretion due to nicotine in cats, the cava pocket method under narcosis being relied on. 0.08 mg. per kg. nicotine was given i.v. (Sugawara)

Adrenaline secretion rate scale is here one-tenth of the common standard in this book, and the time scale is taken ten times as large as the common.

Response to an intravenous application of nicotine is a somewhat rapid and large increase in the blood sugar concentration in a short interval of time after the injection, though not very significant, while on the other hand remarkably quick and large changes set in the arterial blood pressure, as if to correspond to the course of the oversecretion of adrenaline by nicotine there, above illustrated in text and figures. Schematically speaking, the arterial pressure falls from 130 mm. Hg. to 30 to 60 mm., which is recorded 10 to 20 seconds after the start of injection. Some seconds further later the pressure abruptly rises until 200–300 mm. or higher, then tends to descend, and finally 5 to 15 minutes later the pre-injection level is completely recovered. It does not descend there further; the latter fact and the preliminary, profound fall, described above, do not occur in the cava pocket experiment under anesthesia.

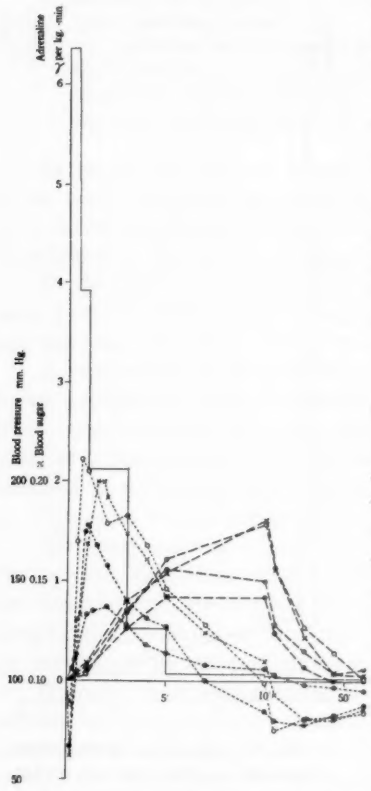

FIG. 37. Blood sugar content and arterial blood pressure in normal as well as in medulli-adrenalectomized dogs, poisoned with nicotine in the i.v. dose of 1 mg. per kg. (S. Kojima, Endo & Nagakura).

Blood sugar—broken line, arterial pressure—dotted line.

Nicotine to normal dogs—light circles; nicotine to medulli-adrenalectomized ones—solid circles; adrenaline hydrochloride saline solution to the dogs so operated on—light circles with point. Nicotine & adrenaline combined to medulli-adrenalectomized dogs—crosses.

Note: Adrenaline scale—1/10 of the common standard; time scale of the first ten minutes—tenfold.

Although there were no material differences in the nicotine hyperglycemia between the normal dog and the doubly splanchnicotomized one, when the blood sugar was measured only a few times in the first interval where the adrenaline secretion is always accelerating extraordinarily,[2,1] a definitely smaller hyperglycemia in comparison with the normal one was always present in the dog deprived of the adrenal medullae, when blood samples were taken several times in the first period.[8] (Fig. 37 & 38)

And an intravenous introduction of adrenaline there, i.e. in the doubly medulli-adrenalectomized dog, in imitation of the adrenaline oversecretion just occurring in the organism on nicotine, can well compensate the shortcoming due to the loss of adrenal medulla re the blood sugar mobilization. Fig. 37 shows the compensatory effect of the adrenaline infusion in respect to the blood sugr content and the blood pressure as well.[8]

In rabbits double splanchnicotomy effects a small diminution in the nicotine hyperglycemia.[9] Double adrenalectomy has no influence at all upon the glycogenic action of nicotine.[7] In the researches quoted above the operation on the adrenal gland or its innervation was carried out several days before the determination of sugar, etc.

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1) Wada, Hirano and Tiba, *Tohoku J. Exp. Med.*, 1938, **33**, 189.

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3) Sugawara, *Ibid.*, 1925, **6**, 430. Literature there.

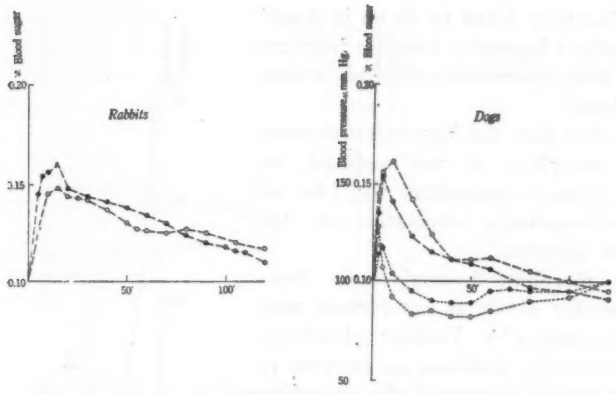

FIG. 38. Blood sugar and blood pressure in nicotine poisoning (1 mg. per kg.) in dogs and rabbits. (Wada)

Blood sugar—broken line, blood pressure—dotted line.

The values in normal individuals—light circles; those in medulli-adrenalectomized dogs respectively in adrenalectomized rabbits—solid circles.

4) Stewart and Rogoff, *J. Pharm. Exp. Ther.*, 19(11–)12, **3**, 379.
5) Sugawara, S. Saito and Nemoto, *Tohoku J. Exp. Med.*, 1927, **9**, p. 207.
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7) Wada, *Tohoku J. Exp. Med.*, 1935, **25**, 14.
8) S. Kojima, Endo and Nagakura, *Ibid.*, 1942, **44**, 95.
9) Inaba and Oikawa, *Ibid.*, 1930, **16**, 169.
* Cf. Wada et al. (l), p. 208.

MORPHINE

Morphine is a hyperadrenalinemic drug. Subcutaneous doses of 10 to 40 mg. per kg. and i.v. doses of 2 to 8 mg. are always effective in this respect, for instance. Fig. 39, A is constructed from five cases of 8 mg. per kg. intravenous application. The hyperadrenalinemia reaches its peak, 0.7 γ per kg.-min., the highest 1.2 γ, about five to ten minutes after the injection; the whole hyperadrenalinemic period covers over two hours and the total amount of oversecretion 30γ per kg.[1,2,3] The secretion is of Type B_1.

Splanchnicotomy totally abolishes this action,[3] which fact is commonly recognized. (Fig. 39, B & C)

When the tolerance to morphine fully develops, that animal loses its hyperadrenalinemic as well as its hyperglycemic ability on morphine.[2]

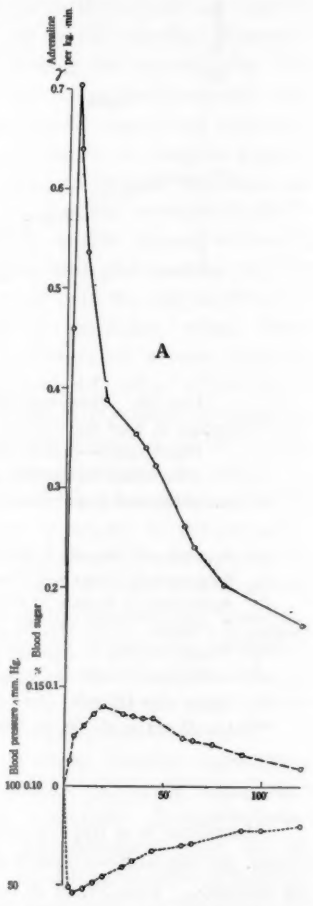
Even in the cava pocket experiment under ether or urethane, the Cleveland physiologists were able to find the hyperadrenalinemic power of morphine

in cats, but they failed to do so in dogs.[4]

Avertin, a hypnotic, does not interfere with the adrenalinemic and glycemic actions of morphine.[5]

The fact that the hyperadrenalinemia due to morphine is not reduced by atropine if any[6] contradicts the idea of the parasympathetic innervation of the adrenaline secretion.[7]

The morphine hyperglycemia develops somewhat slowly in comparison with the adrenalinemia.[1,2] Double splanchnicotomy practically abolishes an increase in the blood sugar content;[3] the morphine hyperglycemia is also entirely of the central origin. (Fig. 39, A, B & C) Brooks et al. apparently would assume it to be the result of a stimulation of the autonomic centres which involve the area of the posterior hypothalamus, cats being experimented on.[8]



In respect to the glycemic event on morphine poisoning, the demedullation of the adrenals behaves quite differently from the splanchnicotomy. The morphine hyperglycemia sets in there in a moderate magnitude and in a similar time course as in the normal individual. Schematically speaking, the highest value is reduced by one-third, and the total amount of excess by two fifths. Tanaka's original figure[9] and Fig. 40, A show this. Similar tendencies are observable in rabbits too.[10] Hypodermical application of the drug given in doses of 3 to 30 mg. per kg., produces a hyperglycemia of 0.17% in normal rabbits, 0.12% in the doubly splanchnicotomized, and 0.12% in the doubly adrenalectomized.

Puzzling it is that Bodo et al. were not able to detect any hyperglycemia on giving morphine to dogs and cats, the adrenals of which were inactivated. They paid attention to the body weight, and moreover, the morphine experiment was carried out on one and the same individual three times, with an interval of ten days between the successive experiments.[11]

The role played by the adrenaline excessively secreted following morphine

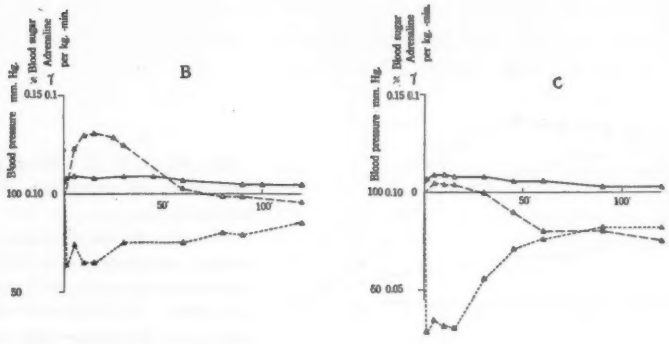


FIG. 39, A, B, C. Adrenaline secretion, blood sugar and blood pressure on 8 mg. per kg. morphine, i. v. injected. (Sato & Ohmi, Wada et al., Sibuta et al.)

"A" is constructed from the experiments on normal dogs, "B" those on unilaterally splanchnicotomized, and "C" those on doubly splanchnicotomized. The values determined in "A" are shown with light circles, and those in "B" & "C" with light triangles.

Adrenaline secretion rate—solid line, blood sugar—broken line, blood pressure—dotted line.

injection in producing hyperglycemia can be readily comprehended from Fig. 40, which was constructed on the data of Tanaka,[9] Sato et al.,[1] Wada et al.[2] and Sibuta et al.[3] .

The hyperglycemia in the medulli-adrenalectomized dog is, as given above, almost similar in shape but definitely smaller in amount in comparison with that in the control animal. And adrenaline saline fluid, intravenously introduced at the rate usually present on morphine administration compensates well this and other shortcomings produced by the de-medullation.

One can see it at a glance in Fig. 2 of Tanaka, reproduced here as Fig. 40, A. In this case the curves are plotted from the actual values, but when all the pre-existing values be taken as 0.1% as in all the other figures in the present treatise,* Fig. 40, B is the resultant plotting. Here the blood sugar increase, however, is comparatively larger than expected, irrespective of whether the adrenaline infusion is done alone or together with morphine. Nevertheless, this is easily and justifiably explained by the fact that the oversecretion of adrenaline taken as the pattern was the largest among the five. At the time of Tanaka's computations we have a single case of 8 mg. morphine per kg. by Sato et al.; the other cases, four in number, were done after Tanaka. The greatest rate of adrenaline secretion there is 1.1 γ per kg.-min., whereas the mean of all the cases is 0.7 γ. The total amount of oversecretion

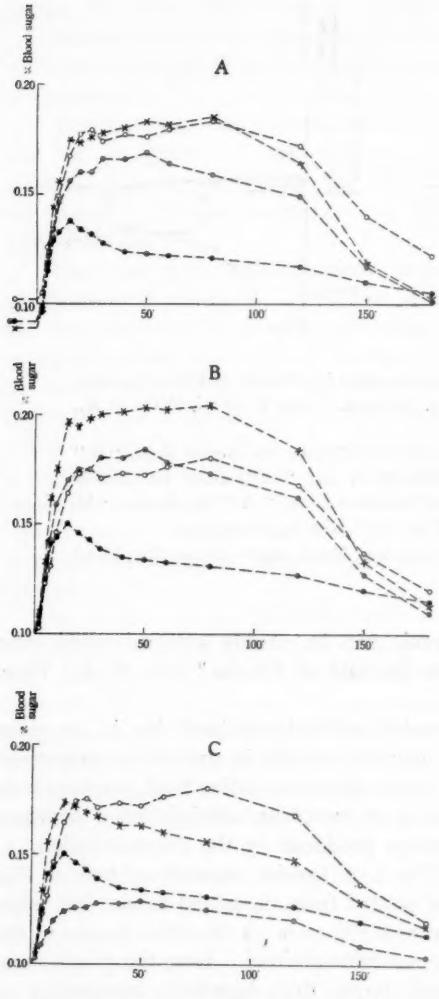


FIG. 40, A, B, C. Blood sugar fluctuation on morphine (8 mg. per kg. i. v.).

Morphine to normal dogs—light circles; morphine to medulli-adrenalectomized ones—solid circles; adrenaline hydrochloride—saline solution to the same—light circles with point; morphine & adrenaline to the same—crosses.

A: Actual values of blood sugar are shown on the ordinate. (Tanaka, *Tohoku J.*, 1936, **30**, Fig. 2 on p. 147).

B: Blood sugar concentration before injection of morphine alone or morphine and adrenaline being taken as 0.1%, as in all other figures in this book.

C: The magnitude of hyperglycemia due to adrenaline infusion is taken as 1/2.9 of that given in "A", and that due to both chemicals as the sum of that by morphine alone and that calculated in the manner just mentioned. Further see the text.

of adrenaline there is in fact 2.9 times the average value, determined thereafter, that example being also involved. The curve obtainable in combining morphine and adrenaline in the medulli-adrenalectomized dog is constructed by combining the new curve of adrenaline infusion and that of morphine alone in the dog so operated on. The results is given in Fig. 40, C. The blood sugar data of Tanaka comprise seven cases. What number is needed for exactly elucidating the physiological role of oversecretion of adrenaline

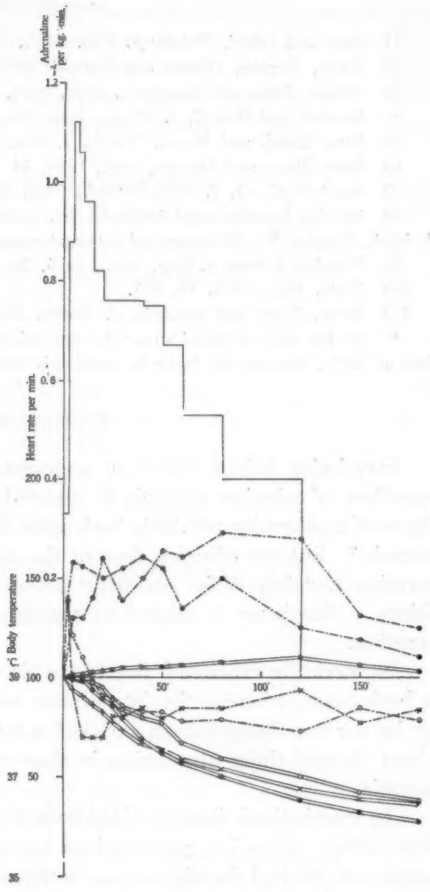


FIG. 41. Role of adrenaline oversecretion in the cardio-acceleration and hypothermia in morphine poisoning. (Tanaka)

Heart rate is expressed by chain line; anal temperature by double line.

The values, modified by morphine in normal dogs are shown by light circles; those in medulli-adrenalectomized ones by solid circles; those by adrenaline infusion carried out as shown in histogram, in the animals operated on by light circles with point, and the same with both drugs in the animals, medulli-adrenalectomized, by crosses.

is suggested on p. 54. These findings verify on the whole the view put forward.

The role played by the oversecretion of adrenaline in morphine intoxication can be detected in the body temperature fall, though not clearly. Remarkable is the finding on the other hand that the combination of morphine and adrenaline in the medulli-adrenalectomized dog produces a similar fluctuation in the pulse rate to that in the normal dog, whereas each of the drugs brings about only the acceleration of the heart beat, as shown in Fig. 41. The actual heart rate is recounted on the base of the body temperature, as is common in this treatise.

References

1) Sato and Ohmi, *Tohoku J. Exp. Med.*, 1933, **21**, 441.
2) Wada, Tanaka, Hirano and Taneiti, *Ibid.*, 1938, **34**, 52.
3) Sibuta, Endo and Nagakura, *Ibid.*, 1949, **50**, 1.
4) Stewart and Rogoff, *J. Pharm. Exp. Ther.*, 1922, **19**, 59.
5) Sato, Degti, and Satow, *Tohoku J. Exp. Med.*, 1935, **25**, 107.
6) Sato, Muto and Hatano, *Ibid.*, 1938, **34**, 393.
7) Kuré et al., *Q. J. Exp. Physiol.*, 1932, **21**, 239.
8) Brooks, Goodwin and Willard, *Am. J. Physiol.*, 1941, **133**, p. 226. See further this book, Chapter VI, Nervous and humoral control of adrenaline secretion.
9) Tanaka, *Tohoku J. Exp. Med.*, 1936, **30**, 123.
10) Tada, *Ibid.*, 1932, **19**, 405.
11) Bodo, Cotui and Benaglia, *J. Pharm. Exp. Ther.*, 1937, **61**, 48.

* In the case of blood sugar the pre-existing value is plotted on the origin, i.e. taken as 0.1%, because the latter is commonly little different therefrom.

Strychnine

Strychnine brings about an augmentation in the adrenaline secretion, regardless of whether narcosis is resorted to or not, and in fact a similar degree of acceleration results in both cases, i.e. with (dogs[1,2] & cats[1]) or without narcosis.[3] But the effective dose of the drug and the absolute rate of oversecretion certainly differ decidedly according to the above mentioned conditions. The latter is related to the largeness of the pre-existing rate of secretion.

Numerically expressed, five to ten times acceleration occurs *ad maximum* in both cases, whereas the intravenous minimum effective dose is 0.25 mg. per kg. for anesthetized dogs, and 0.01 to 0.015 mg. per kg. for unanesthetized. About twenty times strychnine is thus needed when the animal is under narcosis.

In anesthetized dogs provided with the cava-pocket 0.25 to 3 mg. strychnine per kg., given i.v., produced an increased excitability for one and a half hours, and reached the highest rate in the adrenaline secretion 10 to 20 minutes after the injection.[2] In non-anesthetized dogs the peak in the oversecretion was 0.1 γ per kg.-min., and the total amount 5 γ per kg. in less than two hours; roughly said: oversecretion of Type C.[3]

A larger dose of strychnine, such as 3 mg. per kg. in anesthetized dogs and 0.1 mg. in non-anesthetized was not capable of having a stronger effect upon the adrenaline secretion.[2,3] And a still larger i.v.-dose, as 4 to 10 mg. per kg. was soon followed by paralysis and a reduction in the adrenaline secretion in cava-pocket experiments.[2]

As will be seen, the action of strychnine is rather small against what might be generally expected.

Splanchnicotomy abolishes the adrenalinemic action of strychnine,[1,2]

while the action is well demonstrable after the section below the cervical cord.[1]

On administrating strychnine some observers proved the paradoxical pupil dilatation in rabbits,[4,5] which is annulled by double splanchnicotomy, whereas some others failed to detect an acceleration of the denervated heart in cats.[6]

Strychnine, i.v. given, in the amount of 0.05 to 2 mg. per kg. produces a reduction in the adrenaline content of adrenals in rabbits, whereas a too large dose of strychnine, as 5 mg. per kg. does not act so; it causes quick death of the animals.[7]

References

1) Stewart and Rogoff, *J. Pharm. Exp. Ther.*, 1919, **13**, 95; *Am. J. Physiol.*, 1919, **48**, 397; *Ibid.*, 1920, **51**, 484.
2) M. Watanabe, *Tohoku J. Exp. Med.*, 1927, **9**, 251.
3) Aomura and Yen, *Ibid.*, 1929, **14**, 93.
4) Schneider, *Biochem. Ztschr.*, 1922, **133**, 373.
5) Shimidzu, *Arch. exp. Path. Pharm.*, 1924, **103**, 62f (Very weak).
6) Harmon and McFall, *J. Pharm. Exp. Ther.*, 1929, **37**, 131.
7) W. Takahashi, *Tohoku J. Exp.*, 1932, **18**, 327.

Caffeine

In the non-anesthetized dog caffeine, given here intravenously, accelerates the adrenaline secretion.[1] Giving 0.1 g. caffeine per kg. increases the rate by 2–6 times, and 0.15 g. by about fifty times, or 2 γ adrenaline per kg.-min. and 40 γ per kg. are attainable. The peak appears a few minutes after injection, and the whole duration of oversecretion lasts over a half hour. Secretion type B_1. Smaller doses, as 0.003–0.007 g. per kg., have no accelerating effect upon the adrenaline secretion, though they are still capalbe of increasing the body temperature.

For all the dogs, three in number, 0.15 g. per kg. caffeine was lethargic. The animals died 90, 30 or 15 minutes after the injection. The amount is the lethal dose of the caffeine group (i.e. diuretine, theophylline too) for the dog, regardless of being narcotized or not.[1,2]

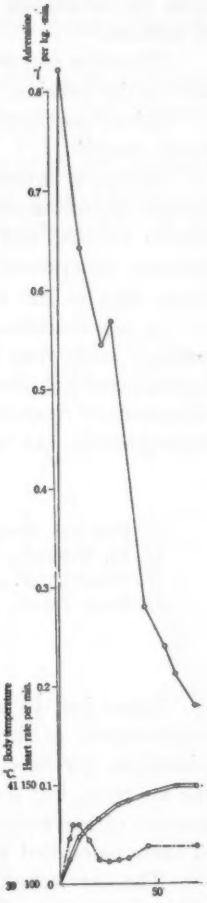


Fig. 42. Adrenaline secretion on caffeine (0.1 to 0.15 g. per kg. i.v.) in normal dogs. (Sato & Aomura)

Adrenaline secretion rate—solid line, heart rate—chain line, anal temperature—double line.

Narcosis acts to lessen to a certain degree the accelerating effect of caffeine upon the adrenaline secretion. Caffeine acts to lessen clearly the deepness of narcosis.[2,1]

Diuretine and theophylline have similar actions re the adrenaline secretion to the caffeine.

Splanchnicotomy abolishes the hyperadrenalinemic power of the caffeine group drugs.[2]

Diuretine hyperglycemia is classical. It remains on a large scale after double splanchnicotomy; the highest content was measured as 0.27% in normal rabbits, and 0.21% after the neurotomy. The amount of hyperglycemia determinable in doubly splanchnicotomized rabbits, is definitely larger than in the fastening diabetes, operation diabetes, etc.[3]

In cats diuretine produces hyperglycemia too, but there are some special features, other than those found in rabbits. That is, while the first period hyperglycemia is almost as same as in rabbits, in cats it is followed by a development of moderate hyperglycemia which lasts for several days. And this hyperglycemia can occur after double splanchnicotomy.[4]

References

1) Sato and Aomura, *Tohoku J. Exp. Med.*, 1929, **13**, 117.
2) M. Watanabe, *Ibid.*, 1928, **10**, 177.
3) Mikami, *Jap. J. Med. Sc., Pharmac.*, 1926, **1**, 121.
4) Fujii, *Tohoku J. Exp. Med.*, 1930, **15**, 285.

Ephedrine

Ephedrine, in an i.v. dose of 30 mg. per kg. body weight, causes an oversecretion of adrenaline in the non-anesthetized dog. The peak, 6 γ adrenaline per kg.-min. (4 examples) was reached about five minutes after the injection, but the plateau was held for one hour or more, and the total amount of oversecretion 40 γ adrenaline per kg. in 100 minutes. The half of these cases died within thirty minutes of intoxication.

The oversecretion of adrenaline is usually accompanied by an elevation of blood pressure. The pulse rate, reckoned on the base of body temperature, remains almost unaltered. Blood sugar varies also sometimes as if to go parallel with the adrenaline secretion, but not always. The body temperature fluctuates almost parallel with the adrenaline secretion.

On the administration of ephedrine a slight excitement occurs, but later is replaced by a depression, and a rapid clotting and sometimes darkness of blood are observed.

Reference

W. Takahashi, Inaba and Wada, *Tohoku J. Exp. Med.*, 1935, **25**, 310.

Lobeline

Lobeline increases the adrenaline secretion and blood sugar. Narcosis does not reduce them.

In the dog, not anesthetized, 1.25–10 mg. per kg. lobeline, subcutaneously given, augments the secretion rate to mean 0.1 γ per kg.-min. and the total amount of oversecretion to 0.8 γ per kg. in two hours.[1] Type B_2.

In the dogs, anesthetized by chloralose, Houssay and Molinelli estimated 10 or 50 times acceleration in the adrenaline secretion on 1 mg. lobeline, i.v. given, the excess being 3 γ per kg.-min. A complete denervation of the adrenals does not suppress this action.[2]

Glycemic action of the drug in rabbits, either intravenously given or subcutaneously, is diminished to a certain degree by double splanchnicotomy, and largely or almost entirely by double adrenalectomy.[3]

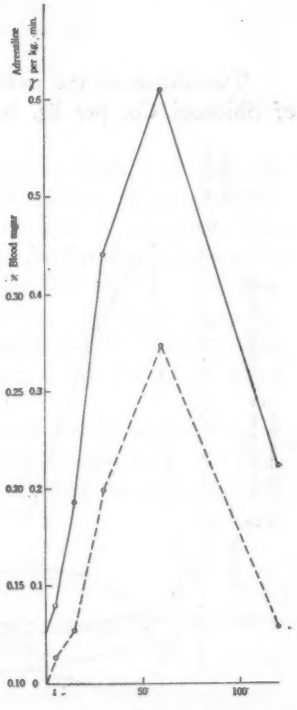


FIG. 43. Adrenaline secretion and blood sugar content after subcutaneous injection of camphor in 2.5 to 5 g. per kg. (Oikawa & T. Inaba)

Adrenaline secretion rate—solid line; blood sugar—broken line.

References

1) E. Inaba, *Tohoku J. Exp. Med.*, 1935, **27**, 348.

2) Houssay and Molinelli, *C. r. Soc. Biol.*, 1925, **93**, 124; *Am. J. Physiol.*, 1926, **76**, 562 ff., 596 ff.

3) Tiba, *Tohoku J. Exp. Med.*, 1938, **33**, 107.

Camphor and Vitacamphor

Camphor increases the adrenaline secretion and the blood sugar in the dog, unanesthetized, not fastened, if it be given subcutaneously in a dose above 1 g. per kg. On that dose slight clinical symptoms and a slight oversecretion of adrenaline are observable.[1] Fig. 43 is composed from two experiments where 2.5 g. and 5 g. camphor per kg. produced severe symptoms, and the animals eventually died. The highest secretion rate was on an average 0.6 γ per kg.-min. and was reached 30 to 60 minutes after the injection. One hour after the injection the rate is apt to decrease. The total oversecretion was 60 γ adrenaline per kg. in two hours duration. Secretion type B_2 is

seen here with this drug and vita-camphor.

Vitacamphor, allo-*p*-oxycamphor, produces in the unanesthetized dog an oversecretion of adrenaline and hyperglycemia on a small scale when 2.4 cc. vitacamphor is intravenously injected. Severe symptoms are elicited, 0.16 γ adrenaline per kg.-min. and an oversecretion of 1.5 γ per kg. in one hour.[2]

REFERENCES

1) Oikawa and Inaba, *Tohoku J. Exp. Med.*, 1930, **16**, 298.
2) Saizyo and Hatano, *Ibid.*, 1936, **29**, 555.

DIGITALIS

Two-thirds of the minimum lethal dose of digitalis (1.5 cc. digitamin of Shionogi Co. per kg. body weight) definitely augments the adrenaline secretion and reduces the blood sugar concentration in the dog, unanesthetized and non-fastened. The development of hyoglycemia is not so rapid and not so profound as in the insulin poisoning. The mean highest rate on 1 cc. digitamin was 0.4 γ adrenaline per kg.-min., and the mean total oversecretion 20 γ per kg. in three hours. (Cf. Fig. 44)

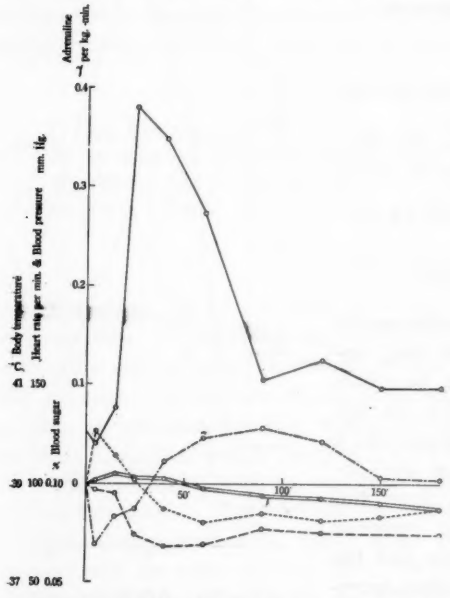


FIG. 44. Adrenaline secretion, etc. on i.v. injection of digitalis in 1 cc. per kg. (Saizyo & Hatano)

Adrenaline secretion rate—solid line, blood sugar—broken line, blood pressure—dotted line, pulse rate—chain line, anal temperature—double line.

A greater dose (1.5 to 4 cc. per kg.) which quickly causes death with deep dyspnoea, is accompanied by a greater adrenaline secretion such as 2.5 to 7 γ per kg.-min.

One-third of the minimum lethal dose is not enough to occasion adrenaline oversecretion, while the pulse rate and the blood sugar decreases undoubtedly.[1]

Whether on applying narcosis this hyperadrenalinemic action of digitalis will be

abolished or not can not be decided by comparing the data of two groups of American authors, who made use of methods somewhat similar in principles on anesthetized cats and dogs. Quite opposite outcomes were yielded there.[2,3]

The findings of Richards and Wood that strophantin accelerates the adrenaline secretion, might be taken to suggest where digitalis acts, and further, the abolishment of the oversecretion of adrenaline by double splanchnicotomy was described there too.[2]

REFERENCES

1) Saizyo and Hatano, *Tohoku J. Exp. Med.*, 1936, **29**, 465.
2) Richards and Wood, *J. Pharm. Exp. Ther.*, 19(14–)15, **6**, 283.
3) Stewart and Rogoff, *Ibid.*, 1919, **13**, 361.

RHODEIN

Rhodein, a glycoside isolated from rhodea japonica Roth, and clinically used as a cardiotonica and diuretica, is also an agent causing hyperadrenalinemia. An injection of 0.8 to 0.9 mg. per kg. rhodein is effective. The peak, mean 0.5 γ adrenaline per kg.-min., ten times acceleration is observable some ten minutes after the i.v. injection, and about 37 γ per kg. adrenaline is secreted in one and a-half hours after the injection. Secretion Type B_2. Smaller doses than the above given are almost ineffective re the adrenaline discharge and other matters, and a larger dose, e.g. 1 mg. per kg. is lethal in ten minutes or thereabouts.

Blood sugar decreases slowly and the lowest, e.g. 0.07% ,appears about 100 minutes after the injection. Pulse rate increases little by little, the peak of about 300 beats per min. being reached two hours after the injection.

REFERENCE

Hatano and Saizyo, *Tohoku J. Exp. Med.*, 1936, **29**, 563.

β-TETRAHYDRONAPHTHYLAMINE

In normal dogs β-tetrahydronaphthylamine produces clinical symptoms similar to those in cats, but in somewhat smaller strength. In intravenous dose of 2 to 5 mg. per kg., it produces an oversecretion of adrenaline, small and short. Commonly two to three times acceleration takes place; in rare cases 10 times. The peak was 0.02 to–0.1 γ adrenaline per kg.-min. and was observed some minutes after the injection; 0.7 γ per kg.-min. only in a single case.[1]

In the dogs provided with the cava pocket preparation under ether,

an acceleration of adrenaline of approximately the same degree as above, i.e. 1.5–15 times was measured when 1 to 3 mg. β-tetra was given. As a matter of course the absolute value of the greatest acceleration is large, i.e. 0.2 to 10 γ per kg.-min. In cats, using the cava pocket preparation, nearly the same degree of acceleration i.e. 2 to 7 times acceleration occurs on i.v. injection of the drug in 1–1.6 mg. per kg., the absolute highest rate being mean 2.5 γ (1 to 4 γ) per kg.-min. Splanchnicotomy totally abolishes this action of β-tetra.[2]

Briefly said, β-tetrahydronaphthylamine has comparatively little effect upon the adrenaline secretion, while it produces an enormous excitment in the animal. The action is not interferred with by the use of narcotics, and is abolished by the splanchnicotomy.[2]

The adrenaline content of the adrenals is made to diminish by β-tetra.[2]

FIG. 45. Adrenaline secretion on i.v. injection of KCl in 30 mg. per kg. (Arai, Oouti & R. Satake)
The determinations in normal dogs—light circles; those in splanchnicotomized ones—light triangles.

REFERENCES

1) Sato and Sugawara, *Tohoku J.. Exp. Med.*, 1930, **16**, 580.
2) Sugawara, *Ibid.*, 1927, **9**, 468.

INORGANIC SALTS

Sodium nitrite, intravenously applied in doses of 20 to 40 mg. per kg. body weight, causes in the dog, unanesthetized, unfastened, an acceleration in the adrenaline secretion. The latter dose is almost a lethal one.

Injection of 30 mg. per kg. sodium nitrite resulted in 0.2 γ adrenaline per kg.-min., two hours' hypersecretion and 15 γ per kg. as the total amount for two cases.

After the injection hyperglycemia of a small magnitude and of a short duration was estimated. In every case the arterial blood pressure descended at once, and the low pressure, say 70 to 80 mm. Hg., was observed for two hours or longer; the heart rate accelerated.[1]

Potassium chloride, intravenously given in doses of 20 to 30 mg. per kg., produces a transitory, rapid increase in the adrenaline secretion rate, the peak being noted as 0.4 to 0.45 γ adrenaline per kg.-min. (the highest 1.2 γ), the duration of oversecretion as

less than one minute and the total amount of excess secretion as 0.3 to 0.4 γ per kg. The secretion is of Type B_2.

Splanchnicotomy reduces it largely: 0.12 γ adrenaline per kg.-min. and 0.08 γ per kg. on 30 mg. KCl per kg.

While a first dose of 20 to 30 mg. KCl per kg. causes a great, but very short acceleration in the adrenaline secretion, as explained above, the second dose, injected thirty minutes or so later, can bring about only a small acceleration, such as one-third or one-fourth of that seen on the first occasion. And if the interval of time between the two injections be made longer, the second acceleration is correspondingly greater.[2]

Ammonium chloride and *magnesium sulphate*, both hypodermically injected in the de-afferented region, produces the paradoxical reaction in cats. It is completely abolished by double splanchnicotmy. The neurotomy is capable of abolishing the hyperglycemia due to ammonium chloride, but effects to reduce the hyperglycemia by another salt only in a small degree.[3]

Both salts do not act in rabbits to reduce the adrenaline content of the adrenals if they are given in sufficient doses, and splanchnicotomy does not change the outcome.[4]

REFERENCES

1) Satake, Sato and Abe, *Tohoku J. Exp. Med.*, 1937, **31**, 416.
2) Arai, Oouti and R. Satake, Will be published.
3) Morita, *Tohoku J. Exp. Med.*, 1922, **3**, 363.
4) Syozi, *Ibid.*, 1937, **30**, 259.

TOXINS

Tetrodotoxin, isolated from the ovary of globe-fish (tetrodon), subcutaneously given, increases the adrenaline secretion in the dog, non-anesthetized, non-fastened. An injection of 0.3 to 0.4 mg. tetrodotoxin caused an acceleration of 2 to 5 times, i.e. as an average 0.1 γ adrenaline per kg.-min., which was reached ten minutes after the injection, and 7 γ per kg. in about two hours.[1] The secretion is of Type B_2.

Tetrodotoxin reduces the adrenaline content of adrenals in rabbits.[2]

Diphtheria toxin was proved in the non-anesthetized dog to have an accelerating influence on the adrenaline secretion when an adequate dose be applied; an intermediate dose increased the secretion while smaller or greater doses had no action as a rule.[3] The greatest rate there was 2 γ per kg.-min., i.e. 20 times acceleration.

REFERENCES

1) Aomura, Yen and Oikawa, *Tohoku J. Exp. Med.*, 1930, **15**, 36.
2) Kimura, *Ibid.*, 1927, **9**, pp. 61–63.
3) M. Watanabe, *Ibid.*, 1928, **10**, 29.

CHAPTER VIII

SECRETION OF SYMPATHOMIMETIC HORMONES AND THEIR PHYSIOLOGICAL ROLE IN CERTAIN FUCTION CHANGES IN THE ORGANISM

CONTENTS: Role of adrenal medulla and other sympathomimetic hormones in regulation of blood sugar — Rate of the heart *in situ* as an indicator of changes in the rate of secretion of adrenaline and other sympathomimetic hormones——Denervated iris for estimating the rate of adrenaline and the like.

THE discovery of the glycemic action of adrenal extract at the very beginning of the present century gave rise to the view that hyperglycemia of the central mechanism is produced exclusively by the oversecretion of adrenaline from the adrenals. Double adrenalectomy annuls its occurrence completely. The outcome, however, was later proved to be not sufficiently correct; a careful investigation usually uncovers the hyperglycemia of a certain manigtude there, even after excluding the adrenals in rabbits, the adrenal medullae in dogs, or the like. The problem still remaining at the present time is how the exclusion effects the magnitude of hyperglycemia of this nature, and what role is played there separately by the increased secretion of adrenaline, and by that of the hormones, orginating from tissues other than the adrenals.

In the experiments on the almost normal dog, given in the preceding chapter, blood sugar was measured simultaneously with the adrenaline secretion in various experimental conditions, and a trial was made in some instances to replace the function of the adrenal medulla by intravenous infusion of adrenaline hydrochloride solution. These data, taken altogether with the outcome from rabbits under similar conditions, which are partly given there also, and partly given here in this chapter, show us a general trend in respect to the question raised above.

Next, denervated organ methods were elaborated first for directly estimating the secretion of adrenaline from the adrenals, chiefly by British as well as American physiologists.[1] However, the reverse is not always true, as later it was elucidated that sympathomimetic hormones are found in tissues other than the adrenals, and act similarly and simultaneously with adrenaline from the adrenals upon denervated organs in experimental conditions such as those at issue. The use of denervated organ methods in fact facilitated the discovery of the sympathomimetic hormones other than the adrenaline.

The heart came in for much research along those lines, particularly in

Boston, Cleveland, and also here. In order to estimate sympathomimetic hormones, it may be mentioned here, that in measuring variations in the rate of the heart, for example the denervated heart, much employed by previous authors, one must take into account constantly the influence of the temperature of the heart itself upon the heart rate *in situ*. Doing this has proved in reality to be very fruitful, as will be illustrated below.

The denervated iris can also be experimented on in an animal without fastening, anesthetizing or evoking any trouble when carrying out the measurement. And the data therefrom are also useful for acknowledging the secretion of adrenaline and other sympathomimetic hormones.

The denervated limb or spleen, for example, were also utilized for measuring the adrenaline secretion, but under conditions much different from the normal physiological. Later investigators[2,3] failed to find any difference in their volume variations due to sensory stimulation before and after exclusion of the medulli-adrenal activity in contrast to the previous workers.[4,5] Such findings correspond to those in the experiments with the denervated heart in the early stage when the method had not at that time been well worked out.

References

1) Early history, Kodama, *Tohoku J. Exp. Med.*, 1923, **4**, 172 ff.
2) Stewart and Rogoff, *Am. J. Physiol.*, 19(22–)23, **63**, 436.
3) F. Watanabe, *Tohoku J. Exp. Med.*, 1935, **27**, 556.
4) Anrep, *J. Physiol.*, 1912(–13), **45**, 318.
5) Pearlman and Vincent, *Endocrinology*, 1919, **3**, 121.

Role of Adrenal Medulla and other Sympathomimetic Hormones in Regulation of Blood Sugar

CONTENTS: Is there any agent which acts to increase the blood sugar only through the splanchnic nerves? —Parts played by splanchnic nerves respectively by adrenal medulla in production of hyperglycemia—Interrelationship between hyperglycemia and oversecretion of adrenaline, simultaneously occurring—Role of oversecretion of adrenaline by a given stimulant upon the blood sugar fluctuation.

A simple approach to the question of how the medulli-adrenal function is essentially concerned in the production of certain experimental hyperglycemia has been tried by many experimentalists of various subjects. A certain stimulation is given once to an individual while it lives wholly normal, and once to another which remains well long after removal of the adrenals (rabbits, etc.) or of the adrenal medullae or the like (dog, cat).

Data obtained under similar experimental conditions are necessary for arriving at a conclusion common to various agencies and various kinds of animals. Procedures which by themselves interfere with the carbohydrate metabolism must be avoided. This measure itself provides in reality the

background for offering the required experimental conditions.

Here, blood samples were collected in rabbits, dogs, and cats from the posterior branch of the ear lobe vein, completely denervated. The cervical sympathetic trunk and the major auricular nerve were cut several days previously, so that the collection of blood could be done not only without anesthetizing, fastening or evoking any pain, but also with ease because of rapid outflow. Such precautions, though seemingly trifling, are necessary for an accurate determination in any study on the blood sugar and the like. The normal blood sugar concentration in these animal species may be taken approximately as 0.1% on an average; as a matter of course as 0.08%, if the method of Dotti[1] be relied upon.

Further noted: rabbits can commonly well survive the double adrenalectomy indefinitely long, similar to normal ones. Such healthy rabbits have considerably enlarged accessories, and accordingly are only a little inferior to the normal in the resistance against several drugs, toxins as acetonitril, nicotine, typhoid toxin, etc.[2] All rabbits have the accessory cortical tissues.[3]

The hyperglycemic data from the rabbits, dogs or cats, normal, doubly splanchnicotomized, deprived of the adrenals and adrenal medullae respectively, without being complicated by other manipulations are summarized in Tabs. XIII & XIV in some detail, and they are again epitomized in Tab. XV. In some dogs the adrenaline secretion experiment was simultaneously determined; they are marked with an asterisk. Some experiments carried on under conditions dissimilar in some ways to the majority of ours are also shown there as the supplement or the control.

Is there any agent which acts to increase the blood sugar only through the splanchnic nerves? There is a certain kind of hyperglycemia, the occurrence of which entirely depends upon the integrity of the splanchnic nerves, called the hyperglycemia of central mechanism, and this is a generally accepted fact even at the present time. Some decades have passed since this observation was first made; meanwhile several methods and precautions for precisely estimating blood sugar and collecting blood specimens have been elaborated step by step.

Some figures in Tabs. XIII, XIV & XV point to the necessity of modifying this view in some way. The maximum hyperglycemia causable by certain agents in doubly splanchnicotomized animals is as small as 0.11% or thereabouts. This exceeds the normal or quiet value only by 0.01%, but it should not be taken as ranging within the limit of error nowadays. That this inference is correct is also clearly illustrated in reality by the fact that in the experiments where as small a hyperglycemia as 0.11% or 0.12% is estimated, the blood sugar concentration before the agent is applied is always exceeded by that collected thereafter if any. Very seldom is one and the same sugar content

discovered before and after an agent is introduced. Such are cases in the experiments with KCl (2 cases in 11 experiments), bleeding (2 in 14), chloretone (2 in 5), morphine (1 in 14) and diphtheria toxin (1 in 10) in rabbits and bleeding (3 in 11) in dogs, while no similar cases were noted in other experiments on asphyxia and histamine in dogs and piqûre, cold-puncture, and heat-puncture +50°C. in rabbits.

The outcome suggests the existence of a portion of the central diabetes, which is brought about through a route or routes other than the splanchnics. There are also some agents known to act directly on the adrenal medulla itself whereby hyperglycemia results. Whether there are agents or not, which produce hyperglycemia through the splanchnic nerves alone is thus open to question. It is of course without question that no hyperglycemia takes place if only a weak stimulation or small dosage, though still effective, be applied there.

TABLE XIII

*Effect of Double Splanchnicotomy upon Hyperglycemia due to Various Agencies.**

(Agencies are arranged within each section approximately in the order adopted in Chapter VII)

Rabbits

Cases where the hyperglycemia is almost annulled or considerably reduced by double splanchnicotomy

KCN: Ohmi, *Tohoku J. Exp. Med.*, 1933, **21**, 107: 2.5 mg. per kg. subc., 0.11% against 0.18% (control animal).

Bleeding: Tachi, *Tohoku J.*, '28, **10**, 96: 1/7 & 1/4 of total blood volume, 0.12% against 0.17% & 0.23%; 1/3–2/5, 0.145% against 0.27%.

Cold-puncture: Morita, *Tohoku J.*, '21, **2**, 403: 0.1–0.12% against 0.26% (operation hyperglycemia being excepted).

Sensory stimulation: Ohmi, *Tohoku J.*, '33, **20**, 498: 0.11% against 0.15%.

Fastening on table: Fujii, *Tohoku J.*, '21, **2**, 9; Hirayama, *Ibid.*, '26, **8**, 37: 0.14% against 0.21%.

Heat-puncture+box temp. 50°C.: Li, *Tohoku J.*, '53, **58**, 11: 0.11% against 0.17%.

Ether: Fujii, *Tohoku J.*, '21, **2**, 169; Tachi & Hirayama, *Ibid.*, '26, **8**, 41: 0.14% against 0.25%.

Remark: In all the cases presented in Tables XIII-XV, the average maximal hyperglycemia is computed from the maximum value in each experiment, while in some papers, particularly in those published more recently and in the foregoing chapter, the whole course of the blood sugar fluctuation is averaged and expressed by the numerals respective figures. Accordingly, the maximum degree of hyperglycemia given in these tables is a little greater than that given in the former chapter, and each assay for one and the same experimental series. The figures for cooling experiments on dogs may be taken as an example thereof: the maximum hyperglycemia in doubly medulli-adrenalectomized dogs and normal animals is given in this chapter as 0.176% against 0.19%, while they are to be taken from Fig. 30, B on p. 92 as 0.163% against 0.175%.

* Needless to say, the subjects here quoted are not all that proved hyperglycemic; Cf. Trendelenburg, Bayer (in Hirsch), Hartman, etc.

Chloretone: Tada, *Tohoku J.*, '31, **17**, 579: 0.1–0.2 g. subc., 0.12% against 0.19%.
Morphine: Tada, *Tohoku J.*, '32, **19**, 405: 3–30 mg. subc., 0.12% against 0.17%; Shen, *J. Biochem.*, '36, **21**, 173: 2 mg. subc., 0.11% against 0.18%
Strychnine: Sakurai, To be published: 0.5 mg. subc., 0.13% against 0.18%.
Diphtheria toxin: Mikami, *Tohoku J.*, '25, **6**, 299: 0.12% against 0.21%.

Cases where the hyperglycemia is reduced by half

Urethane: Tachi & Hirayama, *Tohoku J.*, '28, **10**, 191: 1 g. subc., 0.15% against 0.2%.
Pilocarpine: Tada, *Tohoku J.*, '33, **20**, 275: 2–6 mg. subc., (0.5–6 mg. for control), 0.13% against 0.155%.
Physostigmine: Tada, *Tohoku J.*, '33, **20**, 275: 0.3–1.0 mg. subc., 0.12% against 0.15%.
Lobeline: Tiba, *Tohoku J.*, '38, **33**, 107: 3.5 mg. i.v., 0.135% against 0.15%, 7 mg. 0.15% against 0.2%, 5 mg. subc., 0.13% against 0.14%., 10 mg. 0.14% against 0.17%, 20 mg. 0.14% against 0.20%.
Ligating renal vessels (operation hyperglycemia being excepted): Morita, *Tohoku J.*, '22, **3**, 226: 0.25% against 0.45% (0.28% on the day before rabbits, operated on, died).

Cases where the hyperglycemia is reduced by a varying lesser degree

Peptone: Kaiwa, *Tohoku J.*, '33, **20**, 365: 0.1–0.3 g. i.v., 0.15% against 0.18%.
Guanidine: Tada, *Tohoku J.*, '32, **19**, 584: 0.25–0.45 g. subc., 0.13% against 0.15%.
Heat-puncture+box temp. 60°C.: Li, *Tohoku J.*, '53, **58**, 11: 0.14% against 0.17%.
Nicotine: T. Inaba & Oikawa, *Tohoku J.*, '30, **16**, 169: 2.5 mg. subc., 0.145% against 0.15%; 5 mg. 0.175% against 0.19%.
Telluronium: Kaiwa, *Tohoku J.*, '32, **20**, 163: 10 mg. subc., 0.14% against 0.16%.

Cases where the hypoglycemia is deepened

Insulin: Ohmi, *Tohoku J.*, '33, **21**, 211: Hypoglycemic phase either develops earlier or deepens.
Guanidine: Tada, *Tohoku J.*, '26, **8**, 37: 0.25–0.35 g. subc., 0.06% against 0.08%.

Dogs

Cases where the hyperglycemia is entirely or almost annulled, or considerably reduced

Guanidine: Tada, *Tohoku J.*, '32, **20**, 1: 0.15–0.275 g. per kg. i.v., no hyperglycemia against 0.12%.
Morphine: Sibuta, Endo & Nagakura, *Tohoku J.*, '49, **50**, 1: 8 mg. i.v., 0.1% against 0.15% (control)*; (cats) Stewart & Rogoff, *Am. J. Physiol.*, '22, **62**, 93 (Tables 1 & 2): 0.12% (13 mg. subc.) against 0.2% (2–20 mg.).
Asphyxia: Sato, Inaba & Takahashi, *Tohoku J.*, '32, **19**, 21: 0.12% against 0.19%;* (cats) Kellaway, *J. Physiol.*, '19 (–20), **53**, 225 ff., hyperglycemia of about 0.3% obtainable before and after double splanchnicotmy in a cat.
Bleeding: Tachi, *Tohoku J.*, '28, **11**, 14: 1/3 of total blood volume, 0.115% against 0.19%.
Histamine: Wada, Fuzii, Sibuta, Sakurai & Li, *Tohoku J.*, '40, **37**, 442*: 1 & 2 mg. i.v., 0.105% against 0.155%.
NH_4Cl: (cats) Morita, *Tohoku J.*, '22, **3**, 363: 0.7 g. subc., 0.12% against 0.33%.

Cases where the hyperglycemia is reduced by half

$MgSO_4$: (cats) Morita, *Tohoku J.*, '22, **3**, 363: 0.6 g. subc., 0.2% against 0.3%.

Cases where the hyperglycemia is reduced by a varying lesser degree

Peptone: Kaiwa, *Tohoku J.*, '33, **20**, 365: 0.05–0.3 g. i.v., 0.18% against 0.2%; Saizyo, *Ibid.*, '36, **30**, 34*: 0.05 & 0.1 g. i.v., 0.18% (2 doubly splanchnicotomized).

Cases where the hyperglycemia is slightly reduced or not reduced at all

Asphyxia: (cats) Kellaway, *J. Physiol.*, '19(–20), **53**, 225 ff. (His outcome also may be included here).

Anaphylactic shock: Ohguri, *Tohoku J.*, '35, **25**, 437: 0.155% against 0.16%.

Acetylcholine: E. Inaba, *Tohoku J.*, '35, **27**, 245; Onti, *Ibid.*, '41, **41**, 102: 15 mg. subc., 0.13% against 0.135%; Wada, et al., To be published, 0.12% against 0.11%.

Pilocarpine: F. Watanabe, *Tohoku J.*, '35, **27**, 390*: 1–4 mg. subc., 0.14% against 0.17%.

Nicotine: F. Watanabe, *Tohoku J.*, '35, **37**, 335*: 1 mg. i.v., 0.16% against 0.16%; Wada, Hirano & Tiba, *Ibid.*, '38, **33**, 189*: 1 mg. i.v., 0.125% against 0.14%.

Diuretine: (cats) Fujii, *Tohoku J.*, '30, **15**, 285: A moderate hyperglycemia lasting for several days develops after double splanchnicotomy on a scale similar to that in cats with intact innervation. This occurs besides operation hyperglycemia.

Cases where the hypoglycemia is accelerated

Insulin: Saizyo, *Tohoku J.*, '36, **30**, 34* and Yen et al., *Ibid.*, '33, **21**, 542* taken together: 5–8 or 10 units i.v., 0.03–0.05% (Saizyo) against 0.035–0.06% (Yen et al).

TABLE XIV

Effect of Double Adrenalectomy in Rabbits or Double Medulli-Adrenalectomy in Dogs upon Hyperglycemia due to Various Agencies

Rabbits

Cases where the hyperglycemia is almost annulled or considerably reduced

Guanidine: Tada, *Tohoku J. Exp. Med.*, 1932, **19**, 584: 0.25–0.4 g. per kg. subc., 0.11% against 0.15%.

Fastening on table: Hirayama, *Tohoku J.*, '26, **8**, 37: 0.13% against 0.21%.

Piqûre diabétique: Stewart & Rogoff, *Am. J. Physiol.*, '18, **46**, 90: 0.2% against 0.3% e.g.; Hatano, *Tohoku J.*, '36, **29**, 455: 0.133% against 0.226%.

Heat-puncture+box temp. 50°C. or 60°C.: Li, *Tohoku J.*, '53, **58**, 11: 50°C. 0.117% against 0.149%; 60°C. 0.138% against 0.172%.

Ether: Tachi & Hirayama, *Tohoku J.*, '26, **8**, 41: 0.14% against 0.25%; Ohmi, *Ibid.*, '33, **21**, 116: On shallow narcosis the difference is small (0.15% against 0.16%), but big on deep narcosis, being 0.16% against 0.3%. Stewart & Rogoff, *Am. J. Physiol.*, '20, **51**, 366: 0.27% on etherization one month after double adrenalectomy.

Chloretone: Tada, *Tohoku J.*, '31, **17**, 579: 0.1–0.2 g. subc., 0.12% against 0.19%.

Nicotine: T. Inaba & Oikawa, *Tohoku J.*, '30, **16**, 169: 0.115% against 0.19%; *v. infra.*

Morphine: Stewart & Rogoff, *Am. J. Physiol.*, '22, **62**, 93: 12–30 mg. subc., 0.12% against 0.2%; Tada, *Tohoku J.* '32, **19**, 405: 3–25 mg. subc. for decapsulated, 4–30 mg. for control, 0.12% against 0.17%.

Cases where the hyperglycemia is reduced by half

Bleeding: Tachi, *Tohoku J.*, '28, **10**, 307 & '28, **10**, 96 (control): 1/7 of total blood volume, 0.14% against 0.17%, 1/4 0.19% against 0.23%, 1/3 0.22% against 0.27%.

Synthaline: Tada, *Tohoku J.*, '32, **19**, 584: 18–25 mg. subc., 0.16% against 0.21%.
Urethane: Tachi & Hirayama, *Tohoku J.*, '28, **10**, 191: 1 g. subc., 0.14% against 0.2%.
Pilocarpine: Tada, *Tohoku J.*, '33, **20**, 275: 2–6 mg. subc., (0.5–6 mg. for control), 0.13% against 0.15%.
Physostigmine: Tada, *Tohoku J.*, '33, **20**, 275: 1 mg. subc., 0.12% against 0.15%.
Strychnine: Sakurai, To be published, 0.5 mg. subc., 0.135% against 0.18%.
Lobeline: Tada, *Tohoku J.*, '38, **33**, 107: 3.5 mg. or 7 mg. i.v., 0.11% against 0.15% or 0.2%.
Telluronium: Kaiwa, *Tohoku J.*, '32, **20**, 163: 10 mg. subc., 0.127% against 0.16%.
NH_4Cl: Satake, *Tohoku J.*, '26, **8**, 26: 0.7 g. subc., 0.27% against 0.36%.

Cases where the hyperglycemia is reduced by a varying lesser degree

Asphyxia: Stewart & Rogoff, *Am. J. Physiol.*, '18, **46**, 90, *Ibid.*, '20, **51**, 366: 0.25% against 0.29% (Calculated from the later assay. If the data in both papers be taken together and those under 0.15% be taken out of account: 0.38% against 0.4%); *Idd., Ibid.*, '22, **62**, 93: 0.14% against 0.2%.
KCN: Ohmi, *Tohoku J.*, '33, **21**, 107: 2.5 mg. subc., 0.16% against 0.18%.
Peptone: Kaiwa, *Tohoku J.*, '33, **21**, 1: 0.1–0.3 g. i.v., 0.17% against 0.19%.
Sensory stimulation: Ohmi, *Tohoku J.*, '33, **20**, 498: 0.155% against 0.154% (In several earlier experiments done on long surviving doubly adrenalectomized rabbits only a hyperglycemia on a much smaller scale is occasioned).
Nicotine: Wada, *Tohoku J.*, '35, **25**, 14: 1 mg. i.v., 0.16% against 0.16%; *v. supra.*

Cases where the hypoglycemia is intensified

Insulin: Tada, *Tohoku J.*, '31, **17**, 588: 3–7 units subc., 0.05% against 0.04%; Ohmi, *Ibid.*, '33, **21**, 221: 0.5 units; 0.04% against 0.05%.
Guanidine: Tada, *Tohoku J.*, '32, **19**, 584: 0.25–0.4 g. subc., 0.06% against 0.08%.
Synthaline: Tada, *Tohoku J.*, '32, **19**, 584: 0.18–25 mg. subc., 0.06% against 0.02%.

Dogs

Cases where the hyperglycemia is reduced by more or less half

Peptone: Kaiwa, *Tohoku J.*, 1933, **21**, 1 to *Id., Ibid.*, '33, **20**, 365: 0.1–0.2 g. per kg. i.v. 0.15% against 0.21%.
Anaphylactic shock: Ohguri, *Tohoku J.*, '35, **25**, 445; 1935, **25**, 504:* 0.14% against 0.16%.
Piqûre diabétique: Kaiwa & Wada, *Tohoku J.*, '34, **23**, 536: 0.125% against 0.17%. (Demedullation done a few hours before the puncture, or one month previously. The value of Yen, Kaiwa & Wada, *Tohoku J.*, '31, **17**, 345,* on normal dogs also 0.16%.)
Morphine: Stewart & Rogoff, *Am. J. Physiol.*, '22, **62**, 93: (dogs) 8.5–40 mg. subc., 0.1% against 0.145%; (cats) 3.6 mg. subc., 0.12% against 0.22% (One adrenal removed, the other denervated or large portion of the latter removed, or demedullated); Tanaka, *Tohoku J.*, '36, **30**, 123: 8 mg. i.v., 0.14% against 0.2%.

Cases where the hyperglycemia is reduced to a degree, reduced slightly, or not at all

Asphyxia: Stewart & Rogoff, *Am. J. Physiol.*, '17, **44**, 543: (cats) 0.19% against 0.2%; *Idd., Ibid.*, '22, **62**, 93: (dogs) 0.15% against 0.19%, cats, 0.17% against 0.18%. (Asphyxia was carried out in succession to morphine experiment in the latter paper.)

Kellaway, *J. Physiol.*, 1919 (–20), **53**, 225 ff.: cats; hyperglycemia of 0.3% obtainable by severe anoxemia in a normal and a doubly adrenalecotomized.

CO: Suzuki, Takahasi & Tamabuti, *Tohoku J.*, '52, **55**, 159: 3% CO-inhalation until convulsions set in, 0.17% against 0.19% .

Bleeding: Sato, Kaiwa & Wada, *Tohoku J.*, '35, **26**, 310: 1/3 total blood volume shed, 0.23% against 0.21% (0.19% in normal dogs in Tachi, *Tohoku J.*, '28, **11**, 14).

Cold bath: Body temp. under 30°C.: Wada, Seo & Abe, *Tohoku J.*, '35, **26**, 381*; Itikawa, *Ibid.*, '35, **27**, 568; Satow, *Ibid.*, '37, **30**, 561: 0.176% against 0.19%.

Nicotine: Wada, *Tohoku J.*, '35, **25**, 14: 1 mg. i.v., 0.166% against 0.18%; S. Kojima, Endo & Nagakura, *Ibid.*, '42, **44**, 95: 1 mg. i.v., 0.115% against 0.166%.

Cases where the hypoglycemia is deepened

Insulin: Saizyo, *Tohoku J.*, '36, **30**, 103: 5 units, 0.04% against 0.055%.

Parts played by splanchnic nerves respectively by adrenal medulla in production of hyperglycemia. In many cases the hyperglycemia found in doubly splanchnicotomized animals was noted commonly as 0.13% to 0.15%, but in other cases it was distinctly large as 0.17%–0.2%: they are bleeding, peptone, anaphylactic shock, cold bath, nicotine, renal vessel ligation; asphyxia too. Between these two group results (0.11%, 0.13–0.15% & 0.17–0.2%) one obviously cannot set up any exact boundary there. Moreover, a similar tendency is also detectable in doubly adrenalectomized rabbits as well as medulli-adrenalectomized dogs on the one hand, and on the other hand the strength of stimuli or the dosage of drugs affect the magnitude of hyperglycemia in normal animals and also animals so operated on; they too, are displayed in Tabs. XIII & XIV. Peptone, anaphylactic shock, renal vessels ligating, and particularly nicotine act to increase largely the blood sugar concentration almost or entirely regardless of whether the splanchnic nerves are remaining intact or not. Either the glycemic impulse through other routes is large enough, or the adrenal medulla is directly affected.

What difference is visible then in respect to the magnitude of hyperglycemia detectable between the splanchnicotomized animals and the adrenalectomized? In the experiments on rabbits the scale of hyperglycemia is definitely larger in the latter than in the former in a small number of cases, i.e. in KCN and bleeding, and only a little larger in the tests with peptone, guanidine, while in the remaining cases, the majority, there are no differences at all: they are fastening, heat-puncture +50°C. & 60°C., chloretone, urethane, pilocarpine, physostigmine, nicotine, morphine, strychnine, lobeline, and telluronium.

Double adrenalectomy and double medulli-adrenalectomy apparently exerts no influence upon the intensity of hyperglycemia induceable by asphyxia, sensory stimulation, nicotine and probably ether. There is little or no deviation in the blood sugar concentration between the control cases and the operated.

TABLE XV

Summary of the Effect of Double Splanchnicotomy and Double Adrenalectomy in Rabbits and Dogs upon Hyperglycemia due to Various Agencies

(Summary of the data presented in two foregoing tables)

Agencies	Rabbits		Dogs	
	Splanchnicotomy	Adrenalectomy	Splanchnicotomy	Medulli-adrenalectomy
Asphyxia		0.25% con 0.29% 0.38% con 0.4%	0.12% con 0.2% cat, Kellaway 0.3% no material diff.	S-R. 0.15% con 0.19% cat, 0.19% con 0.19% 0.17% con 0.18% cat, Kellaway, no difference
CO				
KCN	0.11% con 0.18%	0.16% con 0.18%		0.17% con 0.19% (1 g. con 3 g.)
Bleeding	1/7-1/4 0.12% con 0.17% & 0.23% 1/3 0.145% con 0.27%	1/7 0.14% con 0.17% 1/4 0.17% con 0.23% 1/3 0.22% con 0.27%	1/3 0.115% con 0.19%	1/3 0.23% con 0.21%
Peptone	0.15% con 0.18%	0.17% con 0.19%	0.18% con 0.2%	0.15% con 0.21%
Histamine			0.105% con 0.155%	
Anaphylactic shock			0.155% con 0.16% (2.1 g. con 3.4 g.)	0.14% con 0.16% (1.9 g. con 3.4 g.)
Insulin	0.04% con 0.06%	0.05% con 0.04% 0.04% con 0.05%	0.03-0.05 con 0.035-0.06%	0.04% con 0.055%
Guanidine	0.13% con 0.15% 0.06% con 0.08%	0.11% con 0.15% 0.06% con 0.08%		Hyperglycemia con 0.12%
Synthaline		0.16% con 0.21% 0.06% con 0.02%		
Sensory stimulation		0.155% con 0.154%		
Fastening	0.14% con 0.21%	0.13% con 0.21%		
Piqûre diabétique		0.13% con 0.23%		0.125% con 0.17% (0.57 g. con 1.98 g.)
Cold bath				0.176% con 0.19% (3.8 g. con 4.3 g.)
Cold-puncture	0.12% con 0.23%			
Heat-puncture +50° +60°	0.11% con 0.17% 0.14% con 0.17%	0.12% con 0.15% 0.14% con 0.17%		
Ether	0.14% con 0.25%	S-R. 0.27% in adrenalectomized rabbits		
Chloretone	0.12% con 0.19%	0.12% con 0.19%		
Urethane	0.15% con 0.2%	0.14% con 0.2%		
Pilocarpine	0.13% con 0.15%	0.13% con 0.15%	0.14% con 0.17% (1.85 g. con 5 g.)	
Physostigmine	0.12% con 0.15%	0.12% con 0.15%		
Nicotine	2.5 mg. 0.145% con 0.15% 5 mg. 0.175% con 0.19%	0.16% con 0.16% (3.86 g. con 3.52 g.)	0.16% con 0.16% (1.69 g. con 1.42 g.) 0.125% con 0.14% (1.18 g. con 0.95 g.)	0.166% con 0.18% (1.9 g. con 1.23 g.) 0.115% con 0.166% (0.77 g. con 1.87 g.)
Morphine	0.12% con 0.17%	0.12 con 0.17%	0.11% con 0.15%	0.14% con 0.2% (5.18 g. con 9.32 g.) S-R. 0.1% con 0.145% cat, S-R. 0.12% con 0.22%
Strychnine	0.13% con 0.18%	0.135% con 0.18%		

Diuretine	Cat, no difference re hypergl. lasting for several days			
Lobeline	i.v. 0.15% con 0.2%	0.15% con 0.2%		
Telluronium	1.14% con 0.16	0.13% con 0.16%		
NH_4Cl		0.27% con 0.36%	cat, 0.12% con 0.33%	
$MgSO_4$			cat, 0.2% con 0.3%	
Diphtheria toxin	0.12% con 0.21%			
Ligating renal vessels	0.25% con 0.45% (or almost no difference)			

Note: The abbreviation "*con*" in the table signifies "*against*." Numerals in parentheses show amounts of excess sugar.

With dogs asphyxia, bleeding, or morphine produces a definitely greater hyperglycemia in the doubly medulli-adrenalectomized in comparison with the splanchnicotomized, but peptone and anaphylactic shock bring about a contradictory outcome, i.e. a smaller hyperglycemia in the former.

If the data in the medulli-adrenalectomized animals alone be considered, the magnitude of hyperglycemia is commonly smaller in the animals deprived of the adrenal medulla in comparison with the controls. Letting out one-third of the total blood volume results in a rather greater hyperglycemia, but the difference is not large enough to be appreciated here. Briefly, the adrenal medulla make some significant contribution towards the increase of blood sugar due to these agencies.

However, it is noteworthy that in several cases, such as asphyxia, sensory stimulation, bleeding, etc. the magnitude of hyperglycemia before and after interference with the medullary function is not significantly different provided their strength be large enough. This circumstance also led the Cleveland physiologists to their wellknown view that such subjects have no accelerating action upon the adrenaline secretion. It harmonizes well with the data they obtained with their cava pocket method.

The important question of the role of adrenaline secretion upon the variation of blood sugar, blood pressure, etc. is a quantitative matter after all, as repeatedly emphasized by Stewart;[4] two main ways of approaching the question are given in their paper with justification, but at that time, the methods and procedures were not advanced, fully accurate, or free from complicacy.

The numerals re the hypoglycemia in insulin and guanidine experiments also indicate the role of splanchnic nerves and adrenal medulla upon the carbohydrate metabolism, though insignificantly.

Interrelationship between hyperglycemia and oversecretion of adrenaline, simultaneously occurring.

Do adrenaline secretion and blood sugar content go hand in hand when they

change? First by comparing the course of the adrenaline secretion and and that of the blood sugar content some relationship between the two can be deduced; in the majority of cases here investigated both subjects fluctuate hand in hand, roughly speaking. This means only that when one is increasing or decreasing the other is doing so generally. They are asphyxia, peptone, anaphylactic shock, the hyperglycemic phase in guanidine poisoning, piqûre diabétique, cold application, heating, physostigmine, pilocarpine, the later period in nicotine and morphine poisoning, ephedrine, lobeline and camphor.

Here it should be noticed that the adrenaline in adrenal blood was in the present investigations commonly attained by means of the rabbit intestine segment, and sometimes simultaneously in parallel by the paradoxical pupil in a cat. The former method yields somewhat larger value in comparison with the glycemic ability.[5]

Further, if the scales of the adrenaline secretion rate and the blood sugar concentration be taken as shown in this book, in many cases apparently they vary almost parallel. In the cases, however, where the adrenaline rate increases very rapidly as in the very short interval following nicotine or morphine intravenous injection, in CO inhalation or acetylcholine administration, it is almost impossible to be assured of parallelism in figures constructed similarly to those above.

Contrary to the above examples, there are a few cases where the blood sugar content diminishes while the adrenaline secretion increases; they are insulin, digitalis and rhodein, and less definitely in the later stage of guanidine.

A relationship quite the reverse is demonstrable when a dog is anesthetized by means of surgical anesthetics. Under ether or chloroform the rate of adrenaline output is largely depressed on the one hand, and the blood sugar content increases largely on the other hand.

Which precedes the other when adrenaline secretion and blood sugar content start to vary? This kind of examination may reveal any causal relationship that might exist between the two. Preceding of the onset of hyperglycemia proves at least the intervention of other factors in addition to the hyperadrenalinemia in bringing about the increase in blood sugar. When the adrenaline secretion augmentation precedes without fail and with a certain interval of time, its significance in causing the hyperglycemia is suggested.

Agents by which the adrenaline secretion invariably increases first, followed a few minutes later by the appearance of hyperglycemia, are anaphylactic shock, guanidine (hyperglycemic phase), physostigmine (subc.), nicotine and morphine. In other cases, asphyxia, piqûre diabétique, cooling, acetylcholine, lobeline and camphor increase both mostly or frequently simultaneously; outside this either adrenaline or sugar precedes the other. The peak of hyperglycemia or hypoglycemia precedes that of hyperadrenalinemia on bleeding, sugar-puncture and insulin.

Total amount of oversecretion of adrenaline and of excess sugar. No definite relationship running through all the cases can be established here, even roughly, between the maximum acceleration in the rate of adrenaline secretion and the maximum content in hyperglycemia. Nicotine and CO-inhalation produce a remarkably accelerated adrenaline secretion, e.g. 10 γ or 5 γ adrenaline per kg.-min., but the blood sugar content determined there is only 0.15% to 0.2%, i.e. not very different results from those obtainable by other agents, as anaphylactic shock, histamine, camphor, etc. The glycemic ability of adrenaline depends not only upon its concentration existing in the organism at every moment, but also upon the duration acting within the organism. With a query the subheading shows, in mind, both the amounts are arranged on trial as follows:

Asphyxia: oversecretion of adrenaline 12 γ per kg.; excess sugar 3 g. per kg. (in 85 minutes). CO: 25 γ adrenaline; 3 g. sugar (90 min.). Bleeding: 100 γ & 8 g. (120 min.). Peptone: 10 γ & 1.5 g. (120 min.) Histamine: 25 γ & 4 g. (70 min.). Anaphylactic shock: 50 γ & 3.5 g. (150 min.). Piqûre: 100–50 γ & 5 g. (120 min.). Cold water into stomach: 5 γ & 0.55 g. (90 min.). Cold bath: 13 γ & 3.5 g. (90 min.). Heating: 5 γ & 2 g. (50 min.). Acetylcholine: 3 γ & 0.35 g. (60 min). Pilocarpine (subc): 10 γ & 5 g. (150 min.). Physostigmine (subc.): 5 γ & 2 g. (150 min.). Nicotine: 25 γ & 1.5 g. (60 min.). Morphine: 25 γ & 6 g. (150 min.). Ephedrine: 40 γ & 1.4 g. (100 min.). Camphor (subc): 50 γ & 11 g. (180 min.). Vitacamphor: 2–3 γ & 1.5 g. (180 min.).

Ratio of both the amounts in the above cases is thus appreciably dissimilar from case to case, yet a certain correlation exists in the experiments under simple conditions. In thirty cases of rabbits,[5,6] which were doubly splanchnicotomized and intravenously injected with adrenaline-saline solution in an approximately uniform rate for 100 minutes, the ratio is computed as 10 γ adrenaline per kg.: 1.5 g. sugar per kg. as the mean. A ratio of 10 γ per kg.: 1 g. per kg. is found for three sets of experiments on doubly splanchnicotomized dogs. The patterns after which adrenaline hydrochloride solution was intravenously applied are more or less similar to each other in the latter sets of experiments on dogs.[7]

Dissimilarities in the ratio are to be reasonably expectable in the other cases inasmuch as very complicated matters are involved there, which no doubt affect the carbohydrate metabolism in various ways, such as secretion of other sympathomimetic hormones, various courses of adrenaline secretion in form, speed and duration, etc. The data displayed above might be examples thereof. Adrenaline infusion done in medulli-adrenalectomized dogs affords another example; the adrenaline infusion was done there in various differing ways.[8]

Role of oversecretion of adrenaline by a given stimulant upon the blood sugar fluctuation.

Then, to test quantitatively the role of oversecretion of adrenaline by a given stimulant upon the blood sugar fluctuation, simultaneously taking

place, it is desirable to attack the problem directly in the following way. First a simultaneous determination of adrenaline secretion and blood sugar content is made in normal dog with proper intervals of time along the whole course of investigation, in fact before and after giving the stimulus. Next, the blood sugar estimation alone is done just as in the manner above described in the dog, normal and medulli-adrenalectomized as well. In the medulli-adrenalectomized dog, the stimulus is given and followed with an intravenous infusion of adrenaline (hydrochloride) solution in amounts which correspond in every moment, as far as that is possible, to the adrenaline secretion by that stimulus in the normal animal. The latter precedure is alone done in the dog so operated on in completing the set of experiments. The latter three sets of experiments need not be carried out in the order mentioned here.

This whole set of experiments was carried out with bleeding, peptone, anaphylactic shock, insuline, piqûre diabétique, cold bath, nicotine and morphine. Infusion of adrenaline alone was not done in the experiments of bleeding and cold bath.

What number of experiments should be made for setting the mode of fluctuation in the adrenaline secretion and the blood sugar content on applying a given agent in a specific dose is referred to elsewhere (Cf. Chapter VII, p. 54).

With each of these agents about a half dozen experiments were run; fewer in some cases. The figures here presented indicate the general trend in spite of the comparative smallness of the number of experiments.

Here the agencies were applied in doses sufficient to produce an abundant secretion of adrenaline, and one and the same dose was used throughout all the experiments of every set. One-third of the total blood volume was shed, peptone was given in about 0.2 g. per kg., insulin in 5 units per kg., nicotine in 1 mg. per kg. and morphine in 8 mg. per kg.; all intravenously applied. The anal temperature 30°C. or lower was registered in a cold bath. An intermediate strength or dose was not tried in particular.

Glycemic reaction of the organism against these agencies can be differentiated into two classes: with morphine, nicotine, piqûre diabétique and cold bath the orthodox glycemic ability develops competely quantitatively. That is, the hyperglycemia causable by these agents is composed from that by the adrenaline oversecretion occurring there, and that by the factors other than the adrenaline from the adrenals but with the same property as the latter re glycemic ability; a simple algebraic sum of both the elements. Otherwise expressed: the deprivation of adrenal medulla reduces the hyperglycemia due to the agent to a certain extent, and the reduction is adequately compensated by the adrenaline hydrochloride saline solution in the manner and amount secreted thereby from the adrenals in the normal dog. With both

factors, agent and adrenaline infusion combined in the medulli-adrenalectomized dog, hyperglycemia exactly the same as in the normal dog is brought about.

The above statement will be further supported by the data re the heart rate variation in the next section.

The results which can be thus numerically expressed would provide the answer, as we see it, to the question posed by the Cleveland physiologists who said: "Even if it were proved that such increases *in the adrenaline secretion* (words in italics are the author's) could be brought about, what physiological significance could they have, since suppression of the adrenaline output produces no recognizable change in the animal?"[9]

Fig. 10, 12, 19, 22, 30, 37, 40, 15, & 17 prove the above statements well.

Here it must be said that the figure for morphine has been modified (Fig. 40, C) by reducing the dose or effect of adrenaline infusion to about one-third of the original (Fig. 40, A). When Tanaka experimented we had as the pattern only an example of adrenaline secretion on 8 mg. morphine; the total amount of oversecretion of adrenaline there is in fact 2.9 times the average value determined thereafter, that example being also involved. The curve obtainable in combining morphine and adrenaline in the medulli-adrenalectomized dog is constructed by combining the new curve of adrenaline infusion and that of morphine alone in the dog so operated on (Fig. 40, C). Further see text on p. 110 ff. in Chapter VII.

Total amount of adrenaline secreted is 25 γ per kg. for morphine (8 mg. i.v. per kg.), and for nictoine (1 mg.) and 50 γ for piqûre; and adrenaline hydrochloride involved in the injection saline fluid is also 25 γ per kg., 24 γ, 50 γ respectively. In the case with insulin, given below, they are 75 γ per kg. and 78 γ respectively. In cold bath experiments much adrenaline hydrochloride (20 γ) was intravenously administered; it was that amount of adrenaline secreted in a dog (No. 5) of Wada et al. It exceeds certainly the average amount of secreted hormone. In consequence, a definitely large hyperglycemia was produced in the medulli-adrenalectomized dog on cooling, accompanied by the adrenaline infusion. The number of experiments of this set is regrettably small.

The unintelligible effect of a combined administration of peptone and adrenaline on the carbohydrate metabolism reminds us at once of the reversal action of adrenaline. There are an enormous number of reports thereon.* We, too, have had a few such experiences.[10,11] The fact that aeration of adrenaline hydrochloride fluid with oxygen is indispensable for the appearance of the reversal, leads one to give as explanation of the data observed in the peptone and anaphylactic shock experiments under such postulation, that peptone or histamine acts in the long run on the adrenaline hydrochloride, whereby the latter is rendered inactive, whereas the adrenaline coming from the adrenals remains totally unaffected. But simply to abolish the action of adrenaline (hydrochloride) is not the "reversal" action. Moreover, the postulation is merely, tentatively presented here.

Insulin acts hypoglycemic and hyperadrenalinemic at the same time.

Apparently this fact contradicts the agents above referred to, but in reality the occurrence of hypoglycemia stimulates centrally the secretion mechanism of adrenaline. This hyperadrenalinemic action comes to light when the blood sugar content falls below 0.06% in normal as well as depancreatized animals. A profound hypoglycemia is a hyperadrenalinemic agent, and its effect acts in turn to increase the blood sugar or to moderate further deepening of hypoglycemia as is readily seen in Fig. 19. Quite different in form, but quite the same in principle.

On this occasion it may be emphasized here, that the fact that the role of adrenaline oversecretion is quantitatively discovered thus with exactness in producing hyperglycemia due to these subjects, proves that the methods of determining the active principle in the adrenal blood, etc., utilized here, stand within the limit available in this kind of experimentation.

These agents thus bring about the ordinary action of adrenaline, and particularly the glycemic action. Then are there visible other ordinary actions of adrenaline at the same time too? Orthodox reaction of adrenaline, secreted and infused as well, is visible in some cases, and the heterodox in other cases, or no regularities can be found.

The arterial blood pressure behaves in nicotine experiments in quite the same manner as in the glycemic matter; however there is this insignificant difference: the nicotine injection increases the blood sugar less than the adrenaline, while the reverse is true re the arterial pressure.

With morphine no trial was run of the blood pressure. Cold bath experiments show quite orthodox action of adrenaline secretion and infusion upon the blood pressure and heart rate, the latter being computed on the base of body temperature, as mentioned elsewhere. The cardiac acceleration on cooling the dog is almost the same in magnitude and course as seen in the control, and the animal lacking the adrenal medulla but injected with adrenaline hydrochloride. Without the injection that kind of animal shows a definitely smaller cardio-acceleration. Ref. Fig. 30.

While nicotine injection occasions a transitory, large cardio-acceleration, the heart beats slowly on injecting adrenaline in as large a concentration as 6 γ per kg.-min., but for a very short duration when the adrenal medulla be removed. In that kind of dog, nicotine or nicotine with adrenaline acts to speed up the pulse, but the rapidity is in order of a normal dog, and an operated dog, both injected with nicotine, and then the latter with nicotine and adrenaline. Otherwise expressed, the effect of combining nicotine and adrenaline is only arithmetical. Effect of adrenaline alone may be the reversing action of the hormone.

A peculiar figure is discovered with morphine. Morphine and adrenaline applied separately to the medulli-adrenalectomized dog plainly accelerate the heart rate, but when combined, the chief outcome is a slow rate

under the base. At the very beginning a transitory acceleration takes place, but this is replaced a few minutes later by bradycardia. This figure harmonizes well with the variations in the pulse rate in the control individual under morphine. See Fig. 41. Combination of two accelerating agencies causes, on the contrary, a reducing action: a heterogenous action of adrenaline!

In respect to the body temperature, adrenaline secreted or infused shows its ordinary action in the experiments with morphine or insulin, though not distinctly. No regular bearing of the adrenal medulla upon the matters under investigation, other than those described herein, was revealed in these researches.

Ordinary glycemic action of adrenaline is also brought about in the experiment with peptone and anaphylactic shock. However, contrary to the above agents, combined application of each of them and adrenaline infusion, done in imitating the adrenaline oversecretion by that agent, does not betray approximately similar figure of hyperglycemia in a normal dog.

Total oversecretion of adrenaline in peptone poisoning (0.2 g. per kg.) and anaphylactic shock is on an average 12 γ and 30 γ per kg. adrenaline respectively. Adrenaline hydrochloride was intravenously injected, saline solution being used as a vehicle, in the splanchnicotomized (peptone) or the medulli-adrenalectomized (anaphylatic shock) dog in a dose of 15 γ per kg. adrenaline and 30 γ alone or combined with the agent. The data are given in Figs. 15 & 17. Throughout both cases, the hyperglycemia brought about in the dog operated on by each agent alone and that by combined application of that agent and adrenaline are of almost the same magnitude and duration, the total amount of excess sugar being 4.2 g. respectively 4 g. per kg. in the experiments of peptone, and 1.6 g. respectively 1.5 g. in those of anaphylactic shock. A definitely larger hyperglycemia is producible by each agent alone in a normal control. Adrenaline infusion causes a larger (in anaphylaxis) or smaller (peptone) hyperglycemia in the medulli-adrenalectomized dog, in comparison with that inducible when that agent is given alone or with adrenaline.

In the medulli-adrenalectomized dog, anaphylactic shock with or without simultaneous adrenaline infusion produces a similar reaction re the body temperature and pulse rate, though the data are not so clear as in the glycemic matter. Pulse rate behaves in peptone intoxication as above, but only in a later stage.

In medulli-adrenalectomized dogs under ether, peptone given with adrenaline acts re the blood sugar and body temparature almost similarly to adrenaline alone, but re the arterial pressure both drugs simply join together. No definite figure was perceivable so far as the pulse rate is concerned.

In conclusion we may add that in bilaterally adrenalectomized rabbits, combination of peptone and adrenaline brings on hyperglycemia of such

a degree as is causable by peptone alone, notwithstanding adrenaline hydrochloride is given to produce by itself hyperglycemia of a greater strength than by peptone alone. There is no quantitative data on how peptone accelerates adrenaline secretion in rabbits.

Quite a different reaction exists there on the other hand between adrenaline secretion and blood sugar in surgical anesthesia, and further on digitalis and rhodein. The anesthetics distinctly suppress the adrenaline secretion, while they act to increase the blood sugar. However, the double adrenalectomy distinctly interferes with the development of ether hyperglycemia in rabbits.[12] The influence becomes evident on a long continuation of narcosis, say, two or three hours and longer. The deeper the narcosis, the earlier the influence of deficiency. Nevertheless, whether or not the high hyperglycemia (0.2–0.3% against 0.16%) in the later stage of profound anesthesia is due to an accelerated secretion of adrenaline is open to question. In simple cava pocket experiments under ether on dogs, no special variations as in rabbits, above referred to, were found in numerous cases extending for several hours, with the longest example of 4 hours.[13]

Cardiacs, as digitalis and rhodein, accelerate adrenaline secretion and at the same time reduce the blood sugar concentration in a manner similar to insulin. In contrast to the latter, no preliminary, transitory hyperglycemic stage appears in these cases, and the development of hypoglycemia is slow and does not reach 0.06%, which is just sufficiently low to set up a hyperadrenalinemic period on insulin. The peak of hyperadrenalinemia precedes the bottom of hypoglycemia, say, by ten to twenty minutes.

Whether or not any causal relationship exists here between hyperadrenalinemia and hypoglycemia is here inferred only by analogy with insulin. In order to ascertain, the cardiacs must be applied first of all to an individual deprived of the adrenal medulla.

Recapitulation.

Of the interrelationship between variations in the adrenaline secretion and those in the blood sugar concentration in the organism, one is justified in classifying them into at least three varieties, generally speaking. First: both matters vary, roughly speaking, hand in hand; and in fact they increase almost simultaneously. A large number of subjects, hitherto investigated, belongs to this category. This is again to be divided into two subspecies.

a. A given agent produces an oversecretion of adrenaline, which acts to increase the blood sugar concentration, quantitatively. Otherwise expressed: exclusion of adrenal medullae diminishes the hyperglycemia on giving that agent, by a certain intensity, and this lack in the hyperglycemia is just compensated by an intravenous administration of adrenaline (hydro-

chloride), in a manner sufficient to compensate the absence of oversecretion of adrenaline. Morphine, nicotine, severe cooling, piqûre diabétique are examples, and insulin also belongs to this category in principle. Diminution in the blood sugar concentration beyond a certain limit acts to stimulate the central mechanism of adrenaline secretion, directly or indirectly.

b. An agent such as peptone, or anaphylactic shock brings about hyperadrenalinemia and hyperglycemia of a certain magnitude and duration in the normal dog. When the adrenal medullae be removed, the agent is capable of bringing on hyperglycemia of a lesser magnitude, and an intravenous injection of adrenaline hydrochloride, in imitating the adrenaline oversecretion by that agent, does almost the same. Strangely enough one never sees any additional effect of the drugs when given together: giving both drugs in the usual, adequate, and proper manner produces hyperglycemia on about the same scale as that produced by the agent alone. No effect is visible there on adding adrenaline hydrochloride, so we are left with the impression that adrenaline hydrochloride behaves here itself re the carbohydrate metabolism quite differently from the adrenaline secreted naturally. Provisionally it might be regarded as the heterogenous action of adrenaline hydrochloride in contrast to the orthodox action which is common, seen occurring with the subjects above, as morphine, nicotine, etc. Further discussion about this matter and a paradoxical outcome seen in the pulse rate variations in morphine poisoning will be given later together with similar phenomena associated with the denervated heart and pupil questions.

Secondly, there are cases where the blood sugar is decreased while the adrenaline secretion is augmented. Insulin, digitalis, and rhodein are examples. Insulin itself acts primarily to diminish the blood sugar content, and when a certain limit is reached, the adrenaline secretion is accelerated as a consequence, which in turn inhibits the deepening of hypoglycemia. The action of adrenaline oversecretion is in this case orthodox. Insulin belongs thus to I class a. The cardiacs are able to lessen the blood sugar concentration, but not so strongly, nor so rapidly as insulin does.

Thirdly, diabetic action of surgical anesthetics such as ether and chloroform, is classical; nevertheless they act to reduce the oversecretion of adrenaline which circumstances cause to take place. What influence is exercised mutually by both variations is as yet obscure.

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Rate of the Heart *in situ* as an Indicator of Changes in the Rate of Adrenaline Secretion and Other Sympathomimetic Hormones

CONTENTS: Sympathins—Consideration of the influence of body temperature changes upon the heart in taking it as an indicator of the secretion of sympathomimetic hormones, i.e. adrenaline, sympathins—Different thresholds of coldness in causing the secretion of sympathins and of adrenaline—Secretion of various kinds of sympathins, i.e. adrenaline, sympathin remot. and sympathin loc., etc., on extreme cooling of the dog by means of cold bath.

The denervated heart *in situ* is one of the much applied and most elaborated preparations for measuring the adrenaline secretion.[1] The advancement in procedures and techniques in fact owe much to the earnest studies and serious discussion of both groups of physiologists, in Boston and Cleveland.[2] But in making inferences from the outcome yielded by such a method, one should not lose sight of the fact that the reverse is not always true.

The Boston group finally also succeeded in seeing an occurrence of a cardio-acceleration even after functional excluding of the adrenaline secretion, but the question whether its suppression produces no recognizable change in the animal, either during a quiet state or in a state of stress as well, still remains open to-day for certain subjects. The solution of the problem gives rise to another question with similar characteristics; but actual investigations are needed for the latter.

That failure of occurrence of any cardio-acceleration after the major operation is due to insufficiencies in nutrition, particularly protein diet, was the explanation given by the Boston group. This view gives rise to several questions about matters concerning the adrenaline secretion and the like; for example, when a definite difference is found between two sets of experimental data re the activities of denervated organ or of organs, generally speaking, before and after excluding the adrenal secretion, the state of nutrition of the animals must be judged with utmost care and fairly evaluated. Feeding of experimental animals in various countries, and in various laboratories is commonly diverse.

Stewart and Rogoff concluded their discussions on the denervated heart with the words: " It is for Doctor Cannon to exclude, if he can, by control experiments, other possible factors in the reaction which he attributes to adrenaline "[1] It is possible, as a matter of course, to search for " other possible factors "; it has been done by Cannon and others one by one, and is still in progress, as shown in Chapter III (sympathins) and Chapter IV (noradrenaline). On the other hand it is still difficult even nowadays to exclude all " other possible factors ". It is rather simple to estimate the total amount of cardio-acceleration on giving an agent under question to the animal while wholly sound, and next after bilateral removal of the adrenal medulla; the difference in both the estimates is to be assumed as the effect of oversecretion of adrenaline there. It is however troublesome to express the cardio-acceleration in terms of adrenaline secretion with exactness.

In passing, it may be noticed here, that it is just enough to remove an adrenal and cut the splanchnic nerves on the other side for excluding the adrenaline secretion of the central mechanism, but this can not substitute the double medulli-adrenalectmy in the above given experimental plan, needless to say.

Sympathins

Of the sympathin theory of Cannon, how it was developed, modified, and criticized inside and outside the Boston school, is quite briefly summarized in Chapters III and IV, with our comments.

At any rate, the postulate of two kinds of sympathin, excitatory and inhibitory sympathins could not only never replace the hypothesis of Langley,[4] on how the adrenaline produces contraction or relaxation in a given set of smooth muscles, but also makes invalid the connotation of the sympathin theory. Nevertheless, it seems to me highly desirable to revive the name of " sympathin " in the sense of a substance or substances which act at the terminal of the structures stimulated by sympathetic nerve impulses, or that come away in the blood stream therefrom, as originally designated by Cannon; but it is to be understood that adrenaline is not included therein.

" Sympathin loc." and " sympathin remot." are used also here; the former designates the sympathin which acts immediately upon the tissue where it is produced, and the latter that which is liberated from a tissue and transported elsewhere in the blood stream, where its action is revealed.

Although, as we see now by the advances made in researches, the connotation of the term " adrenaline " can no longer be considered explicit or precise, all sympathomimetic substances, or ability from the adrenal medulla can still be pertinently expressed by that term.

Such namings are of course conceptional; accordingly they would not be futile even when the time comes that all active principles are to be ex-

pressed quantitatively in chemical terms universally. Even of the adrenal extract, there is much room for furthering investigations along this line of thought. To express the amount of sympathomimetic materials or principles in the adrenal gland and blood by the term "adrenaline" is adherence to usage kept up for half a century, though its connotation is no more exactly correct, strictly speaking, nowadays.

Consideration of the influence of body temperature changes upon the heart rate in taking it as an indicator of the rate of secretion of sympathomimetic hormones, i.e., adrenaline, sympathins.

In estimating the sympathin secretion by means of the denervated heart or of the heart, generally speaking, the influence of the body temperature upon the latter must necessarily be taken into consideration in the first place. This consideration has not been properly appreciated hitherto, even when cooling or heating has been applied to the animal.

A small number among subjects, investigated here, causes a definite, though small, increase or decrease in the body temperature of dogs by 1 to 2°C. Pilocarpine, physostigmine, caffeine and ephedrine are hyperthermic in effect while nicotine, morphine and ether act hypothermic. Etherization extending for 2 to 7 hours reduces it by 3°C.[5] Piqûre diabétique also often induces a reduction such as 2°C., but sometimes only an insignificant fluctuation.

Cooling or heating can alter the body temperature extensively in contrast to the above examples.

Now, the effect of alteration of temperature upon the rate of the isolated heart has been sought by references: the data re dogs from Synder,[6] cats from Knowlton and Starling,[7] and rabbits from Feldman and Clark.[8] Incidentally, references of this kind are very few in number.[9]

From the experimental data of Synder the temperature coefficients of the heart rate of dogs are arranged as follows. The rate of the isolated dog heart at 39°C. is taken here as 100 beats per min.

Temperature	42.0	41.0	40.0	39.0	38.0	37.0	36.0	35.0	34.0	33.0	32.0	31.0	30.0	29.0	28.0	27.0
Coefficient	75	79	85	100	106	114	120	133	147	154	169	178	185	200	222	232

In practice the coefficient is, as a matter of course, more minutely reckoned, i.e. decimally to tenths of a degree, as follows:

39°.0	38.9	38.8	38.7	38.6	38.5	38.4	38.3	38.2	38.1	degrees
100	101	101	102	102	103	104	104	105	105	

At a given temperature the heart with normal activity in the normal medium would beat at the rate given by formula $100 \times \frac{100}{a}$, where a is the co-efficient for that temperature. For

example, if the heart beats at the rate of 55 beats per min. at 30°C., it would beat at 39° at a rate of $55 \times \frac{185}{100} = 100$ beats, per min.; i.e. the heart has the normal rate.

By applying this formula the inclined line of Synder can be shifted to a straight line, the abscissa, as the accompanying Fig. 46 shows. A heart-beat rate taking place actually in an isolated heart, a denervated heart or the normal heart *in situ* can be calculated by this measure and plotted on the figure. Any deviation from the abscissa indicates then a variation in the heart rhythm, produced by a factor or factors other than the temperature, and its magnitude is readily comparable one with the other, and in reality in terms of total variation; further it may to converted into adrenaline or the like.

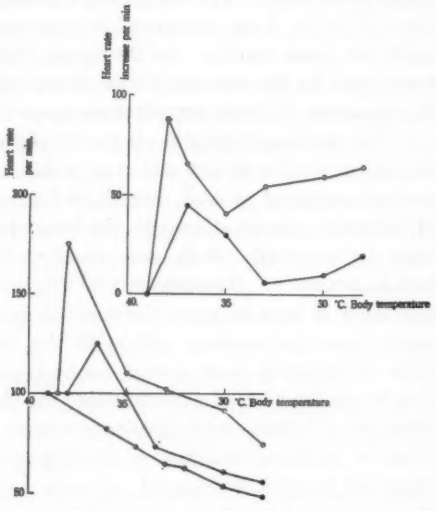

FIG. 46. Rates of denervated heart on cooling normal dogs and medulli-adrenalectomized ones. (Wada & Fuzii)—How to eliminate the direct influence of heart temperature change itself from the change in the heart rhythm which is brought about in the long run, on applying the cold to the animal body.

Heart rate in normal dogs—light circles; that in medull-adrenalectomized ones—solid circles.

Left figure: Actual rate and the Synder line (light circles with point).

Right figure: The rate, recalculated by having shifted the Synder line on the abscissa.

Different thresholds of coldness in causing the secretion of sympathins and of adrenaline.

A small descent of body temperature, such as 0.5 to 2°C. is almost incapable of evoking any oversecretion of adrenaline from the adrenals. Only 0.02 γ adrenaline per kg.-min. is determinable there. The cold is applied to dogs with the lumbar route preparation by means of a rubber coat doubly layered and filled with cold water, or by placing the animal's body in direct contact with an ice block.[10]

And the introduction of ice water into the stomach through a gastric fistula, prepared beforehand, in an amount sufficient to cause heat-debt of about two to three Calories per kg. body weight, brings about as small an acceleration of adrenaline secretion as 0.1 γ per kg.-min.[11,16]

The latter method of causing the coldness was applied to the dog with the completely denervated heart. It was then found that a heat debt of

2.5 to 3 Cal. per kg. accelerates the pulse frequency *ad maximum* by 20 to 40 beats per min., i.e. by 10 to 40 per cent of the initial. The peak is recorded twenty minutes after the water introduction. Demedullation of the adrenals does not materially modify the cardio-acceleration in respect to the degree or the duration.[12] Fig. 28, B is an example which reproduces the actual numerals as such. The body temperature fall does not exceed two degrees.[11] Figs. 27 & 28, A are composed of two sets of experiments,[14,12] which yielded quite the same results. In the figures the pulse rate is as usual in the book rearranged in the manner above given, in order to eliminate the influence of variations of body temperature upon the heart rate.

The cardio-acceleration is practically almost the same in the early period of cooling, say for twenty minutes, in the control individual and in the medulli-adrenalectomized as well, but these latter animals have a cardio-acceleration of definitely greater degree in the later period, and at the same time a lower body temperature. Both are causally related. The body temperature fall acts to set up an oversecretion of sympathin. The fact that the cardio-acceleration is here of quite the same degree before and after the medulli-adrenalectomy harmonizes well with the findings re the adrenaline secretion from the adrenals in so a weak cooling, given above. It may be here added, that in the later set of experiments[11] the denervated pupil was simultaneously observed. Before medulli-adrenalectomy some paradoxical dilatation took place on applying coldness, in the degree reported above, but after the operation the paradox was traced out only with difficulty, in striking contrast to the denervated heart reaction. The events on this reaction are referred to in the next section. A further note may be added: the heart was denervated completely in a three-stage operation; the upper thoracic sympathetic chains on both sides from the stellate ganglion to the 8th thoracic ganglion were removed, the cardiac branches of the left vagus including the recurrent nerve severed, and the right vagosympathetic trunk severed in the neck.[11,12]

Cannon was the first to apply cold to an animal (cats) with the denervated heart; at that time however the excluding of the adrenaline secretion annulled entirely or almost the cardio-acceleration on cooling.[14] If the animal be fed properly no negative results like this would occur, a later experience in Boston.[14]

Nevertheless there were generally still some differences remaining in both groups of investigators (Boston and Cleveland), in respect to the magnitude of reaction remaining after excluding the adrenaline secretion.

Introduction of cold water, with a heat-debt of 2.5 to 3.5 Cal. per kg. induces in dogs[11,15] and cats[16], both provided with the stomach fistula, a body temperature fall by 1 to 2 degrees, and a hyperglycemia of a small magnitude such as 0.11%. In one set of experiments[11] an adrenaline secretion of insignificant degree was measured simultaneously. On an average, the total

amount of oversecretion of adrenaline and of excess sugar there are 3 γ per kg. and 0.55 g. per kg. respectively. Cold bath experiments, where an extensive low anal temperature resulted, gave the figures 13 γ per kg. and 3 g. per kg. The ratio of both factors, adrenaline and sugar, is not markedly different in either case of coldness, weak or strong. This consideration makes it unfair to deny completely the possibility of a causal relation existing between adrenaline oversecretion and blood sugar increase in the cold water into stomach-experiment.

At the same time, the minimum effective dose of adrenaline (hydrochloride) on sustained infusion for two minutes for accelerting the denervated heart is about 0.2 to 0.3 γ per kg.-min.[17,18] This is about two times as large as the maximum rate attainable on the weak cooling as the cause a fall of 1 to 2 degrees in the body temperature. If the infusion be continued for 5 minutes with 0.25 γ adrenaline hydrochloride per kg.-min., the rate of the heart with the intact innervation increases definitely by 30 beats per min. in the medulli-adrenalectomized dog.[17] An acceleration in the secretion rate of adrenaline, estimated in the gastric fistula experiment is thus presumably just sufficient for producing an increase in the blood sugar concentration, the rate of the denervated heart and the arterial pressure. The denervated heart in the dog is accelerated by such a cooling by approximately 20 beats, and in fact on the same scale, regardless of whether the adrenal medulla is interfered with or not, as mentioned above.

If all the evidences and inferences presented above be taken altogether, the acceleration of the denervated heart is produced chiefly by means of the sympathin, when the dog is weakly cooled to cause a fall of 1°C. or a little more in the body temperature. Regrettably we did not attempt to fix the limit of cooling where the sympathin only is secreting, while the adrenal medulla is still at rest. The above works were carried out in fact for elucidating whether the medulli-adrenalectomy reduces or not the acceleration of the denervated heart on cooling from the stomach.

In passing it may be noted here again that even when cold water be introduced through the stomach fistula into the stomach, emotional disturbances can be evoked occasionally in animals. Male cats are apt to suffer therefrom.[16] This tendency in male cats has long been known,[19] as related elsewhere.

Further, the following facts may be added in passing: cooling of the animal body does not prolong the action of adrenaline (hydrochloride), intravenously administered,[20] and exclusion of the adrenals, adrenal medullae and splanchnic nerves do not modify the glycemic effect of adrenaline hydrochoride, hypodermically applied.[21]

Secretion of various kinds of sympathins, i.e. adrenaline, sympathin remot. and sympathin loc., etc., on extreme cooling of the dog by means of a cold bath.

The apparently antagonistic but virtually synergistic studies, particularly with the denervated heart rate of both schools, Boston and Cleveland, led the way to the discovery of sympathin of Cannon in the initial sense. Inasmuch as Cannon's later publications rather seriously tangle, regrettably, the connotation of the word, we see the reason why some writers propose to abandon its use.

Nevertheless, determinations of the heart rate in the dog, extensively cooled, enable one to estimate amounts of adrenalne, sympathin loc. and sympathin remot. In doing this the estimation of the heart rate is to be done with the intact innervations as well as with the complete denervation, and in fact while the adrenal medullae remain intact or are removed.

Some words may be spent by way of precaution on the methods and procedures here used. Superfluous it is to say that all the experiments were done without narcotizing the dog, as is common in all the experiments presented in this book. The dog was immersed by force in a cold bath until the anal temperature descended under 30°C. Cold puncture proves about 30°C. as the upper limit for causing the cold diabetes by serial estimation through the whole period of blood sugar.[23] The adrenaline secretion from the adrenals is accelerated in a cold bath in parallel with the blood sugar. Further, the denervation of the heart, here quoted, was done in addition to the operation above described, the sympathetic nerves were reduced by bilateral removal of the lower abdominal sympathetic chains including five ganglia and by severance of the hepatic nerves.[23]

Now two sets of experiments, one of which was carried out on the dog with the normal heart by Satow[24] and another on the dog with the denervated heart by Wada and Fuzii,[23] are put together. The dogs were all cooled extremely, and the cooling was done before and after the medulli-adrenalectomy; the heart rates are recalculated here on the base of the temperature of the heart, i.e. here of the anal temperature. Figs. 47–50 are constructed in order to show at one glance the influence of various sympathins upon the heart in terms of pulse frequency, the body temperature being taken on the abscissa and the recalculated heart rate on the ordinate.

For illustration: the mean heart rates actually seen in a set of experiments[24] are given in Fig. 46, left, on p. 143., together with the Synder line. They are in fact transformed to Fig. 46, right, according to the principle and manner described there.

Referring to Fig. 47 the left side figure shows the variations in the heart rate when the dog is cooled in the bath causing the body temperature to descend, while the right side indicates the body temperature recovering when the animal is taken out of the bath.

Curves A show pulse rates in the dogs with the intact innervation of heart, and A_1 those while the adrenal medullae are still intact, and A_2 those

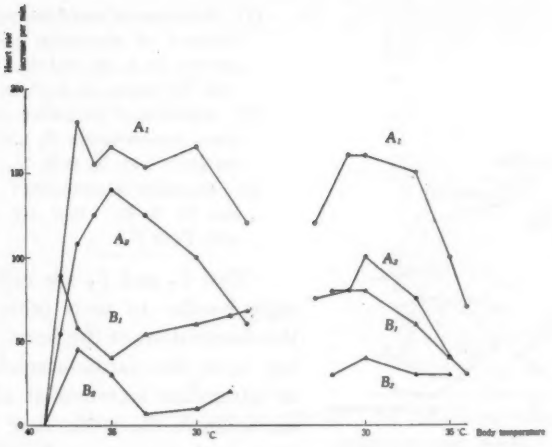

FIG. 47. Heart rate of dogs in cold bath, the rate being recalculated on the base of body temperature, and the direct influence of body temperature upon the heart rate eliminated by the use of Synder line.

A_1 & A_2—Dogs with the heart intactly innervated.

B_1 & B_2—Dogs with the denervated heart.

A_1 & B_1—Dogs with intact adrenals.

A_2 & B_2—Dogs with demedullated adrenals. (Wada et al., Satow)

Further see the text.

after the medullae are removed. Curves B are those from the the dogs with completely denervated heart, B_1 and B_2 are those before and after medulli-adrenalectomy.

These curves, A_1, A_2, B_1 & B_2, show accordingly the heart acceleration due respectively to:

A_1 (dog, wholly normal): decrease in acetylcholine secretion, secretion of sympathin loc. and sympathin remot. plus adrenaline

A_2 (heart intactly innervated, adrenal medullae removed): decrease in acetylcholine secretion, and secretion of sympathin loc. plus sympathin remot.

B_1 (heart denervated, adrenal medullae intact): sympathin remot. plus adrenaline

B_2 (heart denervated, adrenal medullae removed): sympathin remot.

N.B. B_1 & B_2 are there given in Fig. 46.

It is to be noted that the operation practised in denervating the innervation of the heart, effects a certain reduction in the secretion of sympathin remot. as a byproduct. Accordingly the variations, represented by B_1 and B_2, must be properly evaluated, in taking this circumstance into account.

From the figures on the left side in Fig. 47, the amount of cardio-acceleration by each factor can be deduced as follows:

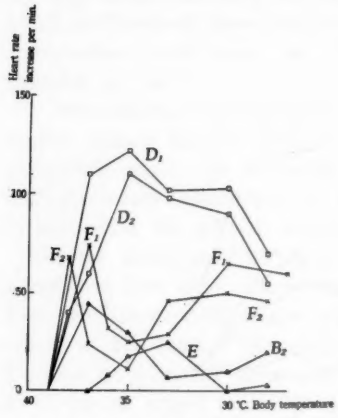

FIG. 48. Heart rate variations due to secretion of adrenaline, sympathins in the cold bath. Body temperatures are taken on the abscissa.

D_1, D_2 indicate cardiac acceleration due to reduction of acetylcholine secretion plus secretion of sympathin loc.

B_2, E indicate cardiac acceleration due to secretion of sympathin remot.

F_1, F_2 indicate secretion of adrenaline. Further see the text.

(1) Reduction of acetylcholine secretion plus secretion of sympathin loc.: by A_1–B_1 and also by A_2–B_2, and they are represented with the curves D_1 & D_2 in Fig. 48.

(2) Secretion of sympathin remot.: by the direct determination B_2 and A_2–D_2; they are given with B_2 & E.

(3) Secretion of adrenaline: by A_1–A_2 and also by B_1–B_2: they are indicated there with F_1 & F_2.

That F_1 and F_2 are rather astonishingly similar to each other shows that the denervation of the heart has no bearing upon the cardio-accelerating action of adrenaline secretion at all. The denervation in the work, here quoted, was done in 4 steps, extending from 2 to 4 months, and the cold bath experiment was carried out soon after the last operation. The medulli-adrenalectomy was done before starting, or during denervation of the heart.

Each one of the three factors, adrenaline, sympathin remot. and acetylcholine-decrease plus sympathin loc., gives in the two figures, arrived at from the different sets of experimentations, a good similarity in the curves of cardio-acceleration, in form and magnitude as well. Only the curves for the sympathin remot. definitely differ from each other, but in the later half period of cooling an approximately similar course of acceleration was noted; here weight must be laid upon the figure, actually observed. On the whole the uniformity of the data would be recognized as acceptable for each factor in such experimental conditions.

Provided the amount of cardio-acceleration, produced while the body temperature decreased from 39°C. to 27°C. be distributed among three kinds of sympathins namely, reduction of acetylcholine plus secretion of sympathin loc., secretion of sympathin remot. and secretion of adrenaline, they are in the ratio of 5 :1 :2. This calculation is not however correct in principle; the abscissa indicates the body temperature, and no time factor is involved in the figure.

FIG. 49

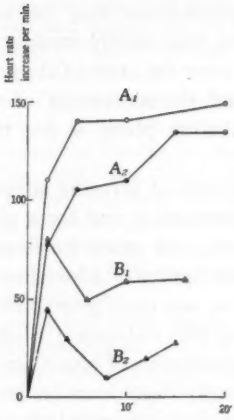

FIG. 50

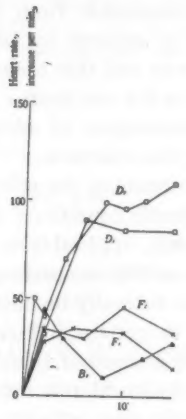

FIG. 49. Heart rate of dogs in cold bath, the time being taken on the abscissa.

Legends are the same as in Fig. 47. Further see the text.

FIG. 50. Heart rate variations due to secretion of adrenaline, and sympathins in the cold bath, the time being taken on the abscissa. Legends the same as in Fig. 48. Further see the text.

In supplementing this shortcoming Figs. 49 & 50, corresponding to the above Figs. 47 & 49, with the same symbols are then drawn up, with heart rates recalculated on the base of body temperature on the ordinate, and with times on the abscissa. The time needed for body temperature fall on a given scale was somewhat different, say 2 to 3 times, depending upon the thickness of the fur. Otherwise the figures of both sets, Figs. 47 & 49 against Figs. 48 & 50, would have much similarity.

And yet the amounts of cardio-acceleration taking place in the first 15 minutes when in the cold bath are to be distributed into — acetylcholine + sympathin loco., sympathin remot. and adrenaline in the ratio of 4:1:2. Almost the same as the ratio above given.

Nevertheless, such a rather complete coincidence may be accidental. The number of experiments are small and individual variations not very small. The outcome shows beyond question the general tendency, however.

From this ratio (4:1:2) and the total amount of cardio-acceleration in the cold bath (17 600 beats) given in Fig. 24, the amount of cardio-acceleration due to the adrenaline secreted there, is calculated as 5 000 beats. This corresponds to the amount of adrenaline secreted there, i.e. 13 γ per kg., then 1 γ adrenaline corresponds to 400 heart beats. If this scheme be applied to the data on Fig. 50, the adrenaline would be liberated in the first 15 minutes

of cold bath in an amount of 1 to 1.2 γ per kg., and this just harmonizes with the value obtainable from Fig. 24. The coincidence may be accidental. The latter is deduced by taking the secretion rate simply straight between the initial rate and that observed 25 minutes after the start of dipping in the cold bath, on the one hand. On the other hand, the occurrence of variations in the concentration of adrenaline, actually taking place, is not taken into account in this inference.

By determining the effect upon the heart rate of infusing adrenaline solution in simple conditions, as in a given concentration and for a given time, a general rule, applicable to various conditions, can never be drawn therefrom, as is readily comprehensible. Even if the figures of adrenaline secretion taking place naturally be imitated as done by us, one must give some due consideration to certain influences in critisizing the outcome. Such are the effect of the amount of vehicles upon the concentration of blood, and whether adrenaline hydrochloride introduced intravenously disappears in the organism with quite the same velocity as when the adrenaline is secreted spontaneously, for example. The former disappears very rapidly, contrary to the previous views, and in fact in almost a similar velocity with the adrenal venous blood, provided they are kept in as natural a condition as possible. By way of precaution this might be added here.[25]

The numerals, e.g. the ratio 4 :1 :2, are confined to the amount of cardio-acceleration in dogs on cooling, strictly speaking. Although double medulli-adrenalectomy and the adrenaline infusion in place of adrenal medulla have similar effects in the cold bath experiment on dogs in respect of the cardio-acceleration, blood sugar, arterial pressure and body temperature, as given in Chapter VII, the numerals can not be applied *a priori* to the matters besides the pulse rate. They must be actually determined. Though secretion of adrenaline and of sympathin remot. be the same in all the cases, the effect of denervating the heart on the heart rate event and that of the liver upon the blood sugar must be determined by each.

And the idea must be applied when experimenting with various agents. While only a few agents are capable of accelerating the normal heart in dogs, even when recalculated on the base of body temperature, on the one hand, there are some agencies which were proved clearly in the Cannon school and others as largely accelerating the denervated heart, on the other hand. The latter are surely worthy of investigation along the line of thought indicated above.

At last it should be remarked that the above finding can never be applied to cats and rabbits,[26] contrary to dogs.

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Denervated Iris for Estimating the Rate of Adrenaline and the Like

The denervated iris method is one of the two which are advocated in Trendelenburg[1] next to the method which allows collection at given times of the adrenal blood from the de-afferented dog, without narcotizing, without fastening, and without giving any pain. These conditions are met in the former two also, where the sympathomimetic ability is auto-assayed. Adrenaline is here estimated together with sympathin.

Is the iris method then to be evaluated as equal to the denervated heart? Interesting it is that the denervated iris can be used to indicate the adrenaline secretion from the adrenals very closely, though not so exactly, while the heart is much influenced by the secretion of sympathin remot., as given in the above section. This is clearly shown in the cooling experiment, for example.

To dogs with the completely denervated iris and heart as well, cold water was introduced into the stomach through the stomach fistula in 3 minutes,

in such a quantity as to cause a heart debt of 3 Cal. per kg. The introduction was done 11 days after the removal of both ganglia, the upper cervical and the ciliary, where the pupil diameter was still remaining *ad minimum* after the operation. In half of the cases the adrenals were intact, and in the other the adrenal medullae removed. The data are illustrated in Fig. 27.[2] Cardiac acceleration there is of quite the same degree and duration in both sets of experiments under identical conditions, while in another previous cases[3] the pupil was not examined. They are dealt with elsewhere in this book (Chapter VII, Fig. 28 on p. 89).

The paradoxical pupil reaction that took place showed averaged magnitude and duration as follows: The maximum width 2.5 mm. (2–3 mm.), the time to reach the maximum 5–10 min. after introduction, and the duration 25 min. while the adrenals were intact, and 0.5 mm., 4–9 min. and about 15 min. respectively after removal of the adrenal medullae.

Thus it is sure that the paradoxical iris reaction is in this case chiefly due to the oversecretion of adrenaline. The determinations are of such a degree as to make it not unreasonable to take the paradox pupil unconditionally as an indicator of the adrenaline secretion, if there are no other agencies which bring about some definite paradox dilatation in the medulli-adrenalectomized individuals. Had the procedures and techniques applied here not been sufficiently exact, such rudimental paradox might have been missed wholly.

By comparing these data with those of Wada et al.,[4] the following facts present themselves: the pupil reaction is first in reaching its acme and soonest in recovering; the shiverings take place almost simultaneously, then the oversecretion of adrenaline follows and subsequently the hyperglycemia. The body temperature falls and particularly the cardio-acceleration reaches the maximum about ten minutes after that of the pupil dilatation. The finding that the pupil reaction reaches its acme about five minutes before the hyperadrenalinemia is at least partly due to the fact that cold water introduced in the experiments on the adrenaline secretion in 6 to 10 minutes, and in those of the paradoxical reactions in 3 minutes, the effect of which comes into sight in the scales of anal temperature fall too, 2°C. against 1°. Some considerations or experimentations are, however, necessary to settle this question finally.

Additional data from dogs profoundly cooled are added here with Fig. 26. They had the ciliary ganglion intact and were immersed into cold water (0.5–6°C. for 15 to 27 min.), consequently the anal temperature fell considerably i.e. by 8°C. in the normal dogs and by 8.5° in the medulli-adrenalectomized. Owing to the large fall the dilatation of the denervated pupil was also on as large a scale as 4.5 mm. *ad maximum* as the mean, which was arrived at after 7 minutes, and for 70 minutes duration. Double medulli-

adrenalectomy reduced the dilatation considerably; 0.5 mm. was the maximum dilatation there.[5]

How an agency acts to dilate the denervated pupil and how it is affected by the exclusion of adrenaline secretion depends upon the variety of agencies. They are classified by Hartman[6] in cats unanesthetized, with the completely denervated pupil, as, from the strongest, asphyxia, cold, pinna stimulation and dog barking; after removing the adrenals asphyxia was still effective and caused a dilatation of 1–3 mm. in 3 cats out of 4, and cold bath brought about a dilatation of 2 mm. in a single case out of 4. This was seconded by Itikawa in the same kind of animal, in which the ciliary ganglion was not taken out, with the figures (Amount of dilatation, 1. adrenals intact, 2. adrenal medullae removed, 3. splanchnics cut) as asphyxia 4 mm.: 1.2:1.45, cold 3 mm.:0.8:0.7, exercise 2.2 mm.:0.4:0.85, pinna stimulation 2 mm.:0.5:0.4, ammonia into nasal cavity 2.4 mm.:0.2:0.5, barking dog 0.3 mm.:0:0. On asphyxia the paradox occurred invariably even after medulli-adrenalectomy or splanchnicotomy; cold acted similarly, and on ammonia often no reaction was observed.

That severe asphyxia is capable of inducing though small the paradox pupillo-dilatation in cats, deprived of adrenals, is well known from the very beginning of this kind of investigation;[8,9] particularly Stewart et al. saw it with readiness[10] in cats after removal of one adrenal and section of nerves of the other. The quoted authors, therefore, drew an inference quite different from the others.

The paradox produced by ether to some extent survived the adrenalectomy. This is explicable on other grounds; the paralytical secretion of saliva is abundantly inducible by etherization.[11]

That the paradox pupillo-reaction appears only on a small scale if the adrenaline secretion be operatively excluded, makes great contrast with the bearing of that operation on the hyperglycemia and cardio-acceleration by a given agent, given in the above two sections. While the sources of sympathin remot. are inevitably restricted to a certain extent on denervating the heart completely, the two other kinds of experiments do not suffer from such a restriction. Accordingly the sympathin remot. which is produced under the influence of a given agent is presumably the same in both kinds of experiments, sugar and pupil, and definitely less in the denervated heart experiment.

An amazing contrast is exhibited in the cold water into stomach-experiment, reproduced in Fig. 27, where the pupillo-dilatation was almost entirely annulled by the medulli-adrenalectomy, in contrast to the cardio-acceleration of quite the same degree and duration before and after that operation. The sympathin remot. secreted there effects, therefore, the reaction of quite a different scale upon both denervated organs, pupil and heart.

Based on works from the Boston school, this remarkable finding can be readily explained by assuming, that on cooling the animal the hepatic sympathin is the single one which is liberated there, and this sympathin has no action at all or an infinitesimal one on the denervated iris. Asphyxia, sensory stimulation, etc. can elicit the paradoxical pupil reaction after excluding the adrenaline secretion, some more, some less than the cooling. Further, one and same mechanism must be taken as playing its role there too; no boundary can be settled in the amounts of residue dilatation. Other nerves such as the lower abdominal sympathetic chains, the hypogastric nerves, the splanchnic nerves, etc. induce a dilatation of the pupil;[12,13] the hepatic nerves may be exceptional[14]

If so, it may be justifiable indeed to take non-occurrence of the paradoxical pupil reaction in the animal deprived of the adrenaline secretion, despite the applying of a certain agent, while there are on the other hand definite functional alterations in denervated organs, as an indication that only hepatic sympathin is secreted there and no other sympathin remot. The denervated pupil reaction can thus be utilized in estimating the secretion of the adrenaline and the hepatic sympathin by measuring it before and after excluding the adrenaline secretion, and determining simultaneously the activity changes of other denervated organ or organs. In doing this the iris must be utilized without interfering with the circular fibre.[14]

Researches along this line of thought are needed and should be carried out in advance, and in fact are needed for confirming the previous data from studies done from certain other angles, and further expansion of the materials wide and full would be advantageous.

In passing it may be added that in a later work with Lissák, Cannon came to see a similar action on the iris and nictitating membrane, both in the cat under dial of the extract of cat liver vessels and pulp (here no action at all), contrary to the former experiment, in which hepatic nerves, cardio-accelerators were electrically stimulated. The substance, extracted in these researches has properties similar to adrenaline.[15]

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13) Bacq, *C. r. Soc. Biol.*, 1933, **112**, 1551.
14) Cannon and Rosenbluth, *Am. J. Physiol.*, 1935, **113**, 251.
15) Cannon and Lissák, *Am. J. Physiol.*, 1939, **125**, 765; Lissák, *Ibid.*, 778; Id., *Ibid.*, 1939, **127**, 263.

Now let us relate briefly here again some figures presented in this book.

The adrenaline oversecretion is proved to exert exactly or quantitatively its orthodox function in the cases of morphine, nicotine, severe cooling, piqûre diabétique and insulin, and the secretion of adrenaline and sympathins is also exactly determinable on cooling the dog by the amount of the cardio-acceleration. These function changes can thus be calculated with mathematical accuracy, figures worked out in relief. However, such figures can be seen at present still in small portions of the physiological kingdom of the secretion of adrenaline and the like, otherwise expressed in the secretion of the hormones and the environments on which the secretion eventually exerts a direct and indirect bearing.

There are however already found some agents, the hyperadrenalinemic action of which does not harmonize well with the above scheme. Peptone, anaphylactic shock and volatile anesthetics are such examples.

Highly desirous it is to search for supplementary data showing quantitative correlation between the secretion of adrenaline and the like and various functional variations and adjustments causable thereby; the integrative action elucidated here might be a pattern. Existence of obstacles in the way is however already shown in the above examples, and the history of serum therapy warns us thereof too, in obedience to the laws of nature.

At the present stage of these investigations, it should be noticed that the terms such as sympathin, sympathin remot., etc., hepatic sympathin, adrenaline-intestine, etc., etc. are not only for present-day use, but also for use when the time comes that we shall be able to express the sympathomimetic ability or substances quantitatively in chemical terms, without limit. Actually there exist in the organism or a portion thereof still no cases where the substances, indicated with the terms above given, are composed with wholly constant percentages of the sympathomimetic substances, chemically pure. The term "adrenaline" is to be used for the pure hormone (first isolated by Takamine, with the composition $C_9H_{13}O_3N$), for the sympathothomimetic principles altogether in the adrenal gland, and also for those in the blood, adrenal and general, as well. (N. B. It is also the patent name of a drug adopted by two pharmaceutic companies.)

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